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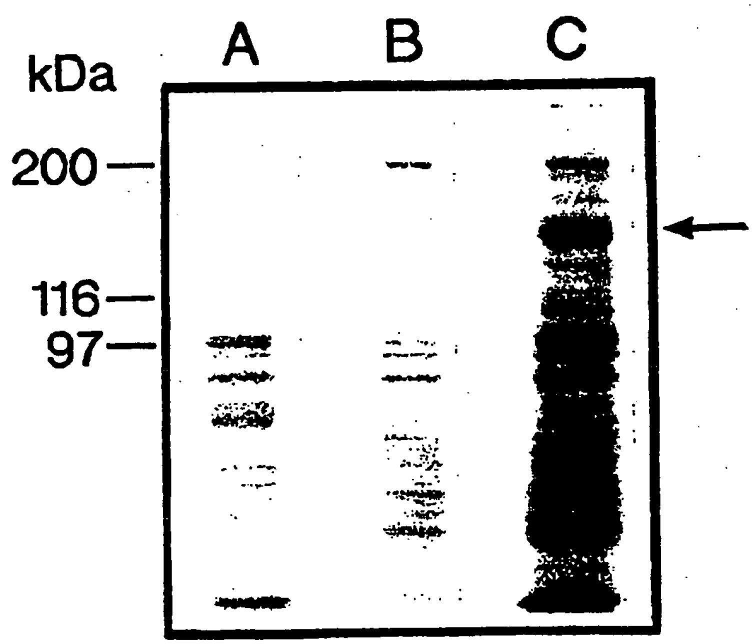
(54) Title: AN INTEGRIN HETERODIMER AND A SUBUNIT THEREOF

(57) Abstract

A recombinant or isolated integrin heterodimer comprising a novel subunit $\alpha 10$ in association with a subunit β is described. The $\alpha 10$ integrin may be purified from bovine chondrocytes on a collagen-type-II affinity column. The integrin or the subunit $\alpha 10$ can be used as marker or target of all types of cells, e.g. of chondrocytes, osteoblasts and fibroblasts. The integrin or subunit $\alpha 10$ thereof can be used as marker or target in different physiological or therapeutic methods. They can also be used as active ingredients in pharmaceutical compositions and vaccines.



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AN INTEGRIN HETERODIMER AND A SUBUNIT THEREOF

FIELD OF THE INVENTION

The present invention relates to a recombinant or isolated integrin heterodimer comprising a subunit α10 and a subunit β, the subunit α10 thereof, homologues and fragments of said integrin and of said subunit α10 having similar biological activity, processes of producing the same, polynucleotides and oligonucleotides encoding the same, vectors and cells comprising the same, binding entities binding specifically to the same, and the use of the same.

BACKGROUND OF THE INVENTION

The integrins are a large family of transmembrane glycoproteins that mediate cell-cell and cell-matrix interactions (1-5). All known members of this superfamily are non-covalently associated heterodimers composed of an α- and a β-subunit. At present, 8 β-subunits (β1-β8) (6) and 16 α-subunits (α1-α9, αv, αM, αL, αX, αIIb, αE and αD) have been characterized (6-21), and these subunits associate to generate more than 20 different integrins. The β1-subunit has been shown to associate with ten different α-subunits, α1-α9 and αv, and to mediate interactions with extracellular matrix proteins such as collagens, laminins and fibronectin. The major collagen binding integrins are α1β1 and α2β1 (22-25). The integrins α3β1 and α9β1 have also been reported to interact with collagen (26,27) although this interaction is not well understood (28). The extracellular N-terminal regions of the α and β integrin subunits are important in the binding of ligands (29,30). The N-terminal region of the α-subunits is composed of a seven-fold repeated sequence (12,31) containing FG and GAP consensus sequences. The repeats are predicted to fold into a β-propeller domain

(32) with the last three or four repeats containing putative divalent cation binding sites. The α-integrin subunits α1, α2, αD, αE, αL, αM and αX contain a ~200 amino acid inserted domain, the I-domain (A-domain), which shows similarity to sequences in von Willebrand factor, cartilage matrix protein and complement factors C2 and B (33,34). The I-domain is localized between the second and third FG-GAP repeats, it contains a metal ion-dependent adhesion site (MIDAS) and it is involved in binding of ligands (35-38).

Chondrocytes, the only type of cells in cartilage, express a number of different integrins including α1β1, α2β1, α3β1, α5β1, α6β1, αvβ3, and αvβ5 (39-41). It has been shown that α1β1 and α2β1 mediate chondrocyte interactions with collagen type II (25) which is one of the major components in cartilage. It has also been shown that α2β1 is a receptor for the cartilage matrix protein chondroadherin (42).

SUMMARY OF THE INVENTION

The present invention relates to a novel collagen type II binding integrin, comprising a subunit α10 in association with a subunit β, especially subunit β1, but also other β-subunits may be contemplated. In preferred embodiments, this integrin has been isolated from human or bovine articular chondrocytes, and human chondrosarcoma cells.

The invention also encompasses integrin homologues of said integrin, isolated from other species, such as bovine integrin heterodimer comprising a subunit α10 in association with a subunit β, preferably β1, as well as homologues isolated from other types of human cells or from cells originating from other species.

The present invention relates in particular to a recombinant or isolated integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, and homologues and or fragments thereof having the

same biological activity.

The invention further relates to a process of producing a recombinant integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or homologues or fragments thereof having similar biological activity, which process comprises the steps of

a) isolating a polynucleotide comprising a nucleotide sequence coding for a integrin subunit α10, or homologues or fragments thereof having similar biological activity,

b) constructing an expression vector comprising the isolated polynucleotide,

c) transforming a host cell with said expression vector,

d) culturing said transformed host cell in a culture medium under conditions suitable for expression of integrin subunit α10, or homologues or fragments thereof having similar biological activity, in said transformed host cell, and, optionally,

e) isolating the integrin subunit α10, or homologues or fragments thereof having the same biological activity, from said transformed host cell or said culture medium.

The integrin subunit α10, or homologues or fragments thereof having the same biological activity, can also be provided by isolation from a cell in which they are naturally present.

The invention also relates to an isolated polynucleotide comprising a nucleotide coding for a integrin subunit α10, or homologues or fragments thereof having similar biological activity, which polynucleotide comprises the nucleotide sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or parts thereof.

The invention further relates to an isolated polynucleotide or oligonucleotide which hybridises to a DNA or RNA encoding an integrin subunit α10, having the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or homologues or fragments thereof, wherein said polyoligo-

nucleotide or oligonucleotide fails to hybridise to a DNA or RNA encoding the integrin subunit α1.

The invention relates in a further aspect to vectors comprising the above polynucleotides, and to cells containing said vectors and cells that have polynucleotides or oligonycleotides as shown in SEQ ID No. 1 or 2 integrated in their genome.

The invention also relates to binding entities having the capability of binding specifically to the integrin subunit α10 or to homologues or fragments thereof, such as proteins, peptides, carbohydrates, lipids, natural ligands, polyclonal antibodies or monoclonal antibodies.

In a further aspect, the invention relates to a recombinant or isolated integrin heterodimer comprising a subunit α10 and a subunit β, in which the subunit α10 comprises the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or homologues or fragments thereof having similar biological activity.

In a preferred embodiment thereof, the subunit β is β1.

The invention also relates to a process of producing a recombinant integrin heterodimer comprising a subunit α10 and a subunit β, in which the subunit α10 comprises the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, which process comprises the steps of

a) isolating one polynucleotide comprising a nucleotide sequence coding for a subunit α10 of an integrin heterodimer and, optionally, another polynucleotide comprising a nucleotide sequence coding for a subunit β of an integrin heterodimer, or for homologues or fragments thereof having similar biological activity,

b) constructing an expression vector comprising said isolated polynucleotide coding for said subunit α10 in combination with an expression vector comprising said isolated nucleotide coding for said subunit β,

c) transforming a host cell with said expression vectors,

d) culturing said transformed host cell in a culture medium under conditions suitable for expression of an integrin heterodimer comprising a subunit α10 and a subunit β, or homologues or fragments thereof similar biological activity, in said transformed host cell, and, optionally,

e) isolating the integrin heterodimer comprising a subunit α10 and a subunit β, or homologues or fragments thereof having the same biological activity, from said transformed host cell or said culture medium.

The integrin heterodimer, or homologues or fragments thereof having similar biological activity, can also be provided by isolation from a cell in which they are naturally present.

The invention further relates to a cell containing a first vector, said first vector comprising a polynucleotide coding for a subunit α10 of an integrin heterodimer, or for homologues or parts thereof having similar biological activity, which polynucleotide comprises the nucleotide sequence shown in SEQ ID No. 1 or SEQ ID No. 2 or parts thereof, and, optionally, a second vector, said second vector comprising a polynucleotide coding for a subunit β of an integrin heterodimer, or for homologues or fragments thereof.

In still another aspect, the invention relates to binding entities having the capability of binding specifically to the integrin heterodimer comprising a subunit α10 and a subunit β, or to homologues or fragments thereof having similar biological activity, preferably wherein the subunit β is β1. Preferred binding entities are proteins, peptides, carbohydrates, lipids, natural ligands, polyclonal antibodies and monoclonal antibodies.

In a further aspect, the invention relates to a fragment of the integrin subunit α10, which fragment is a peptide chosen from the group comprising peptides of

the cytoplasmic domain, the I-domain and the spliced domain.

In one embodiment, said fragment is a peptide comprising the amino acid sequence KLGFFAHKKIPEEEKREEKLEQ.

In another embodiment, said fragment comprises the amino acid sequence from about amino acid no. 952 to about amino acid no. 986 of SEQ ID No. 1.

In a further embodiment, said fragment comprises the amino acid sequence from about amino acid No. 140 to about amino acid No. 337 in SEQ ID No. 1.

Another embodiment of the invention relates to a polynucleotide or oligonucleotide coding for a fragment of the human integrin subunit α10. In one embodiment this polynucleotide of oligonucleotide codes for a fragment which is a peptide chosen from the group comprising peptides of the cytoplasmic domain, the I-domain and the spliced domain. In further embodiments the polynucleotide or oligonucleotide codes for the fragments defined above.

The invention also relates to binding entities having the capability of binding specifically to a fragment of the integrin subunit α10 as defined above.

The invention also relates to a process of using an integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or an integrin heterodimer comprising said subunit α10 and a subunit β, or a homologue or fragment of said integrin or subunit having similar biological activity, as a marker or target molecule of cells or tissues expressing said integrin subunit α10, which cells or tissues are of animal including human origin.

In an embodiment of this process the fragment is a peptide chosen from the group comprising peptides of the cytoplasmic domain, the I-domain and the spliced domain.

In further embodiments of said process the fragment is a peptide comprising the amino acid sequence KLGFFAHKKIPEEEKREEKLEQ, or a fragment comprising the amino acid sequence from about amino acid No. 952 to

about amino acid No. 986 of SEQ ID No. 1, or a fragment comprising the amino acid sequence from about amino acid no. 140 to about amino acid no. 337 of SEQ ID no. 1.

The subunit β is preferably β1. The cells are preferably chosen from the group comprising chondrocytes, smooth muscle cells, endothelial cells, osteoblasts and fibroblasts.

Said process may be used during pathological conditions involving said subunit α10, such as pathological conditions comprising damage of cartilage, or comprising trauma, rheumatoid arthritis and osteoarthritis.

Said process may be used for detecting the formation of cartilage during embryonal development, or for detecting physiological or therapeutic reparation of cartilage.

Said process may also be used for selection and analysis, or for sorting, isolating or purification of chondrocytes.

A further embodiment of said process is a process for detecting regeneration of cartilage or chondrocytes during transplantation of cartilage or chondrocytes.

A still further embodiment of said process is a process for in vitro studies of differentiation of chondrocytes.

The invention also comprises a process of using binding entities having the capability of binding specifically to an integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or an integrin heterodimer comprising said subunit α10 and a subunit β, or to homologues or fragments thereof having similar biological activity, as markers or target molecules of cells or tissues expressing said integrin subunit α10, which cells or tissues are of animal including human origin.

The fragment in said process may be a peptide chosen from the group comprising peptides of the cytoplasmic domain, the I-domain and the spliced domain. In preferred embodiments said fragment is a peptide comprising the

amino acid sequence KLGFFAHKKIPEEEKREEKLEQ, or a fragment comprising the amino acid sequence from about amino acid No. 952 to about amino acid No. 986 of SEQ ID No. 1, or a fragment comprising the amino acid sequence from about amino acid No. 140 to about amino acid no. 337 of SEQ ID No. 1.

The process may also be used for detecting the presence of an integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or of an integrin heterodimer comprising said subunit α10 and a subunit β, or of homologues or fragments thereof having similar biological activity.

In a further embodiment said process is a process for determining the differentiation-state of cells during embryonic development, angiogenesis, or development of cancer.

In a still further embodiment this process is a process for detecting the presence of an integrin subunit α10, or of a homologue or fragment of said integrin subunit having similar biological activity, on cells, whereby a polynucleotide or oligonucleotide chosen from the group comprising a polynucleotide or oligonucleotide chosen from the nucelotide sequence shown in SEQ ID No. 1 is used as a marker under hybridisation conditions wherein said polynucleotide or oligonucleotide fails to hybridise to a DNA or RNA encoding an integrin subunit α1. Said cells may be chosen from the group comprising chondrocytes, smooth muscle cells, endothelial cells, osteoblasts and fibroblasts. Said integrin fragment may be a peptide chosen from the group comprising peptides of the cytoplasmic domain, the I-domain and the spliced domain, such as a peptide comprising the amino acid sequence KLGFFAHKKIPEEEKREEKLEQ, or a fragment comprising the amino acid sequence from about amino acid no. 952 to about amino acid no. 986 of SEQ ID No. 1, or a fragment comprising the amino acid sequence from about amino acid No. 140 to about amino acid no. 337 of SEQ ID No. 1.

In a still further embodiment the process is a process for determining the differentiation-state of cells during development, in pathological conditions, in tissue regeneration or in therapeutic and physiological reparation of cartilage. The pathological conditions may be any pathological conditions involving the integrin subunit α10, such as rheumatoid arthritis, osteoarthrosis or cancer. The cells may be chosen from the group comprising chondrocytes, smooth muscle cells, endothelial cells, osteoblasts and fibroblasts.

The invention also relates to a process for determining the differentiation-state of cells during development, in pathological conditions, in tissue regeneration and in therapeutic and physiological reparation of cartilage, whereby a polynucleotide or oligonucleotide chosen from the nucleotide sequence shown in SEQ ID No. 1 is used as a marker under hybridisation conditions wherein said polynucleotide or oligonucleotide fails to hybridise to a DNA or RNA encoding an integrin subunit α1. Embodiments of this aspect comprise a process, whereby said polynucleotide or oligonucleotide is a polynucleotide or oligonucleotide coding for a peptide chosen from the group comprising peptides of the cytoplasmic domain, the I-domain and the spliced domain, such as a polynucleotide or oligonucleotide coding for a peptide comprising the amino acid sequence KLGFFAHKKIPEEEKREEKLEQ, or comprising the amino acid sequence from about amino acid No. 952 to about amino acid no. 986 of SEQ ID No. 1, or the amino acid sequence from about amino acid No. 140 to about amino acid No. 337 of SEQ ID No. 1. Said pathological conditions may be any pathological conditions involving the integrin subunit α10, such as rheumatoid arthritis, osteoarthrosis or cancer, or atherosclerosis or inflammation. Said cells may be chosen from the group comprising chondrocytes, smooth muscle cells, endothelial cells, osteoblasts and fibroblasts.

In a further aspect the invention relates to a pharmaceutical composition comprising as an active ingredient a pharmaceutical agent or an antibody which is capable of using an integrin heterodimer comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit α10 having similar biological activity, as a target molecule. An embodiment of said pharmaceutical composition is intended for use in stimulating, inhibiting or blocking the formation of cartilage, bone or blood vessels. A further embodiment comprises a pharmaceutical composition for use in preventing adhesion between tendon/ligaments and the surrounding tissue after infection, inflammation and after surgical intervention where adhesion impairs the function of the tissue.

The invention is also related to a vaccine comprising as an active ingredient an integrin heterodimer comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit α10, or DNA or RNA coding for said integrin subunit α10.

A further aspect of the invention is related to the use of the integrin subunit α10 as defined above as a marker or target in transplantation of cartilage or chondrocytes.

A still further aspect of the invention is related to a method of using binding entities having the capability of binding specifically to an integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or an integrin heterodimer comprising said subunit α10 and a subunit β, or to homologues or fragments thereof having similar biological activity, for promoting adhesion of chondrocytes and/or osteoblasts to surfaces of implants to stimulate osseointegration.

The invention is also related to the use of an integrin subunit α10 or an integrin heterodimer comprising said subunit α10 and a subunit β as a target for anti-

adhesive drugs or molecules in tendon, ligament, skeletal muscle or other tissues where adhesion impairs the function of the tissue.

The invention also relates to a method of stimulating, inhibiting or blocking the formation of cartilage or bone, comprising administration to a subject a suitable amount of a pharmaceutical agent or an antibody which is capable of using an integrin heterodimer comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit α10 having similar biological activity, as a target molecule.

In another embodiment the invention is related to a method of preventing adhesion between tendon/ligaments and the surrounding tissue after infection, inflammation and after surgical intervention where adhesion impairs the function of the tissue, comprising administration to a subject a suitable amount of a pharmaceutical agent or an antibody which is capable of using a integrin heterodimer comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit α10 having similar biological activity, as a target molecule.

The invention also relates to a method of stimulating extracellular matrix synthesis and repair by activation or blockage of an integrin heterodimer comprising a subunit α10 and a subunit β, or of the subunit α10 thereof, or of a homologue or fragment of said integrin or subunit α10 having similar biological activity.

In a further aspect the invention relates to a method of in vitro detecting the presence of integrin binding entities, comprising interaction of an integrin heterodimer comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit, with a sample, thereby causing said integrin, subunit α10, or homologue or fragment thereof having similar biological activity, to modulate

the binding to its natural ligand or other integrin binding proteins present in said sample.

The invention also relates to a method of in vitro studying consequences of the interaction of a human heterodimer integrin comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit, with an integrin binding entity and thereby initiate a cellular reaction. Said consequences may be measured as alterations in cellular functions.

A still further aspect of the inventions relates to a method of using DNA or RNA encoding an integrin subunit α10 or homologues or fragments thereof as a molecular target. In an embodiment of this aspect, a polynucleotide or oligonucleotide hybridises to the DNA or RNA encoding an integrin subunit α10 or homologues or fragments thereof, whereby said polynucleotide or oligonucleotide fails to hybridise to a DNA or RNA encoding en integrin subunit α1.

The invention also relates to a method of using a human heterodimer integrin comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit, or a DNA or RNA encoding an integrin subunit α10 or homologues or fragments thereof, as a marker or target molecule during angiogenesis.

BRIEF DESCRIPTION OF THE FIGURES

Fig.1 Affinity purification of the α10 integrin subunit on collagen type II-Sepharose.

Fig. 2. Amino acid sequences of peptides from the bovine α10 integrin subunit.

Fig. 3a. Affinitypurification and immunoprecipitation of the integrin subunit α10 from bovine chondrocytes.

Fig. 3b. Affinitypurification and immunoprecipitation of the integrin subunit α10 from human chondrocytes.

Fig. 3c. Affinitypurification and immunoprecipitation of the integrin subunit α10 from human chondrosarcoma cells.

Fig. 4. A 900 bp PCR-fragment corresponding to the bovine integrin subunit α10

Fig. 5. Schematic map of the three overlapping α10 clones.

Fig. 6. Nucleotide sequence and deduced amino acid sequence of the human α10 integrin subunit.

Fig. 7. Northern blot of integrin α10 mRNA.

Fig. 8 Immunoprecipitation of the α10 integrin subunit from human chondrocytes using antibodies against the cytoplasmic domain of α10 (a). Western blot of the α10 associated β-chain (b).

Fig. 9. Immunostaining of α10 integrin in human articular cartilage.

Fig. 10 Immunostaining of α10 integrin in 3 day mouse limb cartilage.

Fig 11. Immunostaining of α10 integrin in 13.5 day mouse embryo.

Fig 12. Hybridisation of α10 mRNA in various human tissues.

Fig. 13 Immunostaining of fascia around tendon (a), skeletal muscle (b) and heart valves (c) in 3 day mouse limb.

Fig. 14. PCR fragments corresponding to α10 integrin subunit from human chondrocytes, human endothelial cells, human fibroblasts and rat tendon.

Fig 15. Partial genomic nucleotide sequence of the human integrin subunit α10.

Fig 16. Upregulation of α10 integrin subunit in chondrocytes cultured in alginate.

Fig 17. Immunoprecipitation of the α10 integrin subunit from human smooth muscle cells

DETAILED DESCRIPTION OF THE INVENTION

The present invention demonstrate that human and

bovine chondrocytes express a novel, collagen type II-binding integrin in the β1-family. An earlier study presented some evidence for that human chondrosarcoma cells also express this integrin (25). Immunoprecipitation experiments using antibodies against the integrin subunit β1 revealed that this novel α-integrin subunit had an apparent molecular weight (M_r) of approximately 160 kDa under reducing conditions, and was slightly larger than the α2 integrin subunit. To isolate this α-subunit collagen type II-binding proteins were affinity purified from bovine chondrocytes. The chondrocyte lysate was first applied to a fibronectin-Sepharose precolumn and the flow through was then applied to a collagen type II-Sepharose column. A protein with M_r of approximately 160 kD was specifically eluted with EDTA from the collagen column but not from the fibronectin column. The M_r of this protein corresponded with the M_r of the unidentified β1-related integrin subunit. The 160 kD protein band was excised from the SDS-PAGE gel, digested with trypsin and the amino acid sequences of the isolated peptides were analysed.

Primers corresponding to isolated peptides amplified a 900 bp PCR-fragment from bovine cDNA which was cloned, sequenced and used for screening of a human articular chondrocyte λZapII cDNA library to obtain the human integrin α-subunit homologue. Two overlapping clones, hc1 and hc2 were isolated, subcloned and sequenced. These clones contained 2/3 of the nucleotide sequence including the 3' end of the cDNA. A third clone which contained the 5'end of the α10 cDNA, was obtained using the RACE technique. Sequence analysis of the 160 kD protein sequence showed that it was a member of the integrin α-subunit family and the protein was named α10.

The deduced amino acid sequence of α10 was found to share the general structure of the integrin α-subunits described in previously published reports (6-21). The large extracellular N-terminal part of α10 contains a

seven-fold repeated sequence which was recently predicted to fold into a β-propeller domain (32). The integrin subunit α10 contains three putative divalent cation-binding sites (DxD/NxD/NxxxD) (53), a single spanning transmembrane domain and a short cytoplasmic domain. In contrast to most α-integrin subunits the cytoplasmic domain of α10 does not contain the conserved sequence KxGFF (R/K) R. The predicted amino acid sequence in α10 is KLGFFAH. Several reports indicate that the integrin cytoplasmic domains are crucial in signal transduction (54) and that membrane-proximal regions of both α- and β-integrin cytoplasmic domains are involved in modulating conformation and affinity state of integrins (55-57). It is suggested that the GFFKR motif in α-chains are important for association of integrin subunits and for transport of the integrin to the plasma membrane (58). The KxGFFKR domain has been shown to interact with the intracellular protein calreticulin (59) and interestingly, calreticulin-null embryonic stem cells are deficient in integrin-mediated cell adhesion (60). It is therefor possible that the sequence KLGFFAH in α10 have a key function in regulating the affinity between α10β1 and matrix proteins.

Integrin α subunits are known to share an overall identity of 20-40% (61). Sequence analysis showed that the α10 subunit is most closely related to the I-domain containing α-subunits with the highest identity to α1 (37%) and α2 (35%). The integrins α1β1 and α2β1 are known receptors for both collagens and laminins (24;62;63) and we have also recently demonstrated that α2β1 interacts with the cartilage matrix protein chondroadherin (42). Since α10β1 was isolated on a collagen type II-Sepharose, we know that collagen type II is a ligand for α10β1. We have also shown by affinity purification experiments that α10β1 interacts with collagen type I but it remains to be seen whether laminin or chondroadherin are also ligands for this integrin.

The α10 associated β-chain migrated as the β1 integrin subunit both under reducing and non-reducing conditions. To verify that the α10 associated β-chain indeed is β1, chondrocyte lysates were immunoprecipitated with antibodies against α10 or β1 followed by Western blot using antibodies against the β1-subunit. These results clearly demonstrated that α10 is a member of the β1-integrin family. However, the possibility that α10 combine also with other β-chains can not be excluded..

A polyclonal peptide antibody raised against the cytoplasmic domain of α10 precipitated two protein bands with M_r of approximately 160 kD (α10) and 125 kD (β1) under reducing conditions. Immunohistochemistry using the α10-antibody showed staining of the chondrocytes in tissue sections of human articular cartilage. The antibody staining was clearly specific since preincubation of the antibody with the α10-peptide completely abolished the staining. Immunohistochemical staining of mouse limb sections from embryonic tissue demonstrated that α10 is upregulated during condensation of the mesenchyme. This indicate that the integrin subunit α10 is important during the formation of cartilage. In 3 day old mice α10 was found to be the dominating collagen binding integrin subunit which point to that α10 has a key function in maintaining normal cartilage functions.

Expression studies on the protein and mRNA level show that the distribution of α10 is rather restrictive. Immunohistochemistry analyses have shown that α10 integrin subunit is mainly expressed in cartilage but it is also found in perichondrium, periosteum, ossification groove of Ranvier, in fascia surrounding tendon and skeletal muscle and in the tendon-like structures in the heart valves. This distribution point to that α10 integrin subunit is present also on fibroblasts and osteoblasts. PCR amplification of cDNA from different cell types revealed the presence of an alternatively spliced α10 integrin subunit. This spliced α10 was domi-

nating in fibroblasts which suggests that α10 in fibroblasts may have a different function compared to α10 present on chondrocytes.

Expression of the integrin subunit α10 was found to decrease when chondrocytes were cultured in monolayer. In contrast, the expression of α10 was found to increase when the cells were cultured in alginate beads. Since the latter culturing model is known to preserve the phenotype of chondrocytes the results suggest that α10 can function as marker for a differentiated chondrocyte.

Adhesion between tendon/ligaments and the surrounding tissue is a well-known problem after infection, injury and after surgical intervention. Adhesion between tendon and tendon sheets impairs the gliding function and cause considerable problems especially during healing of tendons in e.g. the hand and fingers leading to functional incapacity. The localisation of the α10 integrin subunit in the fascia of tendon and skeletal muscle makes α10 a possible target for drugs and molecules with anti-adhesive properties that could prevent impairment of the function of tendon/ligament. The integrin subunit α10 can also be a target for drugs or molecules with anti-adhesive properties in other tissues where adhesion is a problem.

EXAMPLES

Example 1

Affinity purification of the α10integrin subunit on collagen type II-Sepharose.

Materials and Methods

Bovine chondrocytes, human chondrocytes or human chondrosarcoma cells were isolated as described earlier [Holmvall et al, Exp Cell Res, 221, 496-503 (1995), Camper et al, JBC, 273, 20383-20389 (1998)]. A Triton X-100 lysate of bovine chondrocytes was applied to a fibronectin-Sepharose precolumn followed by a collagen

type II-Sepharose column and the integrin subunit α10 was eluted from the collagen type II-column by EDTA (Camper et al, JBC, 273, 20383-20389 (1998). The eluted proteins were precipitated by methanol/chloroform, separated by SDS-PAGE under reducing conditions and stained with Coomassie blue. (Camper et al, JBC, 273, 20383-20389 (1998). Peptides from the α10 protein band were isolated by in-gel digestion with a trypsin and phase liquid chromatography and sequenced by Edman degradation (Camper et al, JBC, 273, 20383-20389 (1998).

Results

Fig 1 shows EDTA-eluted proteins from the fibronectin-Sepharose (A), flow-through from the collagen type II-Sepharose column (B) and EDTA-eluted proteins from the collagen type II-Sepharose (C). The α10 integrin subunit (160 kDa) which was specifically eluted from the collagen type II column is indicated with an arrow. Figure 2 shows the amino acid sequences of six peptides that were isolated from the bovine integrin subunit α10. Figures 3 a, b, and c show that the α10 integrin subunit is present on bovine chondrocytes (3a), human chondrocytes (3b) and human chondrosarcoma cells (3c). The affinity for collagen type II, the coprecipitation with β1-integrin subunit and the molecular weight of 160 kDa under reducing conditions identify the α10 integrin subunit on the different cells. These results show that α10 can be isolated from chondrocytes and from chondrosarcoma cells.

Example 2

Amplification of PCR fragment corresponding to bovine α10 integrin subunit.

Materials and methods

The degenerate primers GAY AAY ACI GCI CAR AC (DNTAQT, forward) and TIA TIS WRT GRT GIG GYT (EPHHSI, reverse) were used in PCR (Camper et al, JBC, 273, 20383-20389 (1998) to amplify the nucleotide sequence corresponding to the bovine peptide 1 (Figure 2). A 900 bp

PCR-fragment was then amplified from bovine cDNA using an internal specific primer TCA GCC TAC ATT CAG TAT (SAYIQY, forward) corresponding to the cloned nucleotide sequence of peptide 1 together with the degenerate primer ICK RTC CCA RTG ICC IGG (PGHWDR, reverse) corresponding to the bovine peptide 2 (Figure2). Mixed bases were used in positions that were twofold degenerate and inosines were used in positions that are three- or fourfold degenerate. mRNA isolation and cDNA synthesis was done as earlier described (Camper et al, JBC, 273, 20383-20389 (1998)). The purified fragment was cloned, purified and sequenced as earlier described (Camper et al, JBC, 273, 20383-20389 (1998)).

Results

The nucleotide sequence of peptide 1 (Figure 2) was obtained by PCR-amplification, cloning and sequencing of bovine cDNA. From this nucleotide sequence an exact primer was designed and applied in PCR-amplification with degenerate primers corresponding to peptides 2-6 (Figure 2). Primers corresponding to peptides 1 and 2 amplified a 900 bp PCR-fragment from bovine cDNA (Figure 4).

Example 3

Cloning and sequence analysis of the human α10 integrin subunit

Material and methods

The cloned 900bp PCR-fragment, corresponding to bovine α10-integrin, was digoxigenin-labelled according to the DIG DNA labelling kit (Boehringer Mannheim) and used as a probe for screening of a human articular chondrocyte λZapII cDNA library (provided by Michael Bayliss, The Royal Veterinary Basic Sciences, London, UK)(52). Positive clones containing the pBluescript SK+ plasmid with the cDNA insert were rescued from the ZAP vector by *in vivo* excision as described in the ZAP-cDNA® synthesis kit (Stratagene). Selected plasmids were purified and

sequenced as described earlier (Camper et al, JBC, 273, 20383-20389 (1998)) using T3, T7 and internal specific primers. To obtain cDNA that encoded the 5' end of α10 we designed the primer AAC TCG TCT TCC AGT GCC ATT CGT GGG (reverse; residue 1254-1280 in α10 cDNA) and used it for rapid amplification of the cDNA 5' end (RACE) as described in the Marathon™ cDNA Amplification kit (Clontech INC., Palo Alto, CA).

Results

Two overlapping clones, hc1 and hc2 (Figure 5), were isolated, subcloned and sequenced. These clones contained 2/3 of the nucleotide sequence including the 3' end of the cDNA. A third clone (race1; Figure 5), which contained the 5'end of the α10 cDNA, was obtained using the RACE technique. From these three overlapping clones of α10 cDNA, 3884 nucleotides were sequenced The nucleotide sequence and deduced amino acid sequence is shown in Figure 6. The sequence contains a 3504-nucleotide open reading frame that is predicted to encode a 1167 amino acid mature protein. The signal peptide cleavage site is marked with an arrow, human homologues to bovine peptide sequences are underlined and the I-domain is boxed. Metal ion binding sites are indicated with a broken underline, potential N-glycosylation sites are indicated by an asterisk and the putative transmembrane domain is double underlined. The normally conserved cytoplasmic sequence is indicated by a dot and dashed broken underline.

Sequence analysis demonstrate that α10 is a member of the integrin α-subunit family.

Example 4

Identification of a clone containing a splice variant of α10

One clone which was isolated from the human chondrocyte library (see Example 3) contained a sequence that was identical to the sequence of α10 integrin subunit except that the nucleotides between nt positions

2942 and 3055 were deleted. The splice variant of α10 was verified in PCR experiment using primers flanking the splice region (see figure 14).

Example 5

Identification of α10 integrin subunit by Northern blot

Material and methods

Bovine chondrocyte mRNA was purified using a QuickPrep®Micro mRNA Purification Kit (Pharmacia Biotech, Uppsala, Sweden), separated on a 1% agarose-formaldehyde gel, transferred to nylon membranes and immobilised by UV crosslinking. cDNA-probes were 32P-labelled with Random Primed DNA Labeling Kit (Boehringer Mannheim). Filters were prehybridised for 2-4 hours at 42°C in 5x SSE, 5x Denharts solution, 0.1 % SDS, 50 µg/ml salmon sperm DNA and 50% formamide and then hybridised over night at 42 °C with the same solution containing the specific probe (0.5-1 x 106 cpm/ml). Specifically bound cDNA-probes were analysed using the phosphoimager system (Fuji). Filters were stripped by washing in 0.1% SDS, for 1 hour at 80°C prior to re-probing. The α10-integrin cDNA-probe was isolated from the race1-containing plasmid using the restriction enzymes BamHI (GIBCO BRL) and NcoI (Boehringer Mannheim). The rat β1-integrin cDNA probe was a kind gift from Staffan Johansson, Uppsala, Sweden.

Results

Northern blot analysis of mRNA from bovine chondrocytes showed that a human α10 cDNA-probe hybridised with a single mRNA of approximately 5.4 kb (Figure 7). As a comparison, a cDNA-probe corresponding to the integrin subunit α1 was used. This cDNA-probe hybridised a mRNA-band of approximately 3.5 kb on the same filter. These results show that a cDNA-probe against α10 can be used to identify the α10 integrin subunit on the mRNA level.

Example 6

Preparation of antibodies against the integrin subunit α10

A peptide corresponding to part of the α10 cytoplasmic domain, Ckkipeeekreekle (see figure 6) was synthesised and conjugated to keyhole limpet hemocyanin (KLH). Rabbits were immunised with the peptide-KLH conjugate to generate antiserum against the integrin subunit α10. Antibodies recognising α10 were affinity purified on an peptide-coupled column (Innovagen AB).

Example 7

Immunoprecipitation of the integrin subunit α10 from chondrocytes

Material and methods

Human chondrocytes were ^{125}I-labelled, lyzed with Triton X-100 and immunoprecipitated as earlier described (Holmvall et al, Exp Cell Res, 221, 496-503 (1995), Camper et al, JBC, 273, 20383-20389 (1998)). Triton X-100 lysates of 125I-labeled human chondrocytes were immunoprecipitated with polyclonal antibodies against the integrin subunits β1, α1, α2, α3 or α10. The immunoprecipitated proteins were separated by SDS-PAGE (4-12%) under non-reducing conditions and visualised using a phosphoimager. Triton X-100 lysates of human chondrocytes immunoprecipitated with α10 or β1 were separated by SDS-PAGE (8%) under non-reducing conditions and analysed by Western blot using the polyclonal β1 antibody and chemiluminescent detection as described in Camper et al, JBC, 273, 20383-20389 (1998).

Results

The polyclonal peptide antibody, raised against the cytoplasmic domain of α10, precipitated two protein bands with Mr of approximately 160 kD (α10) and 125 kD (β1) under reducing conditions. The α10 associated β-chain migrated as the β1 integrin subunit (Figure 8a). To verify that the α10 associated β-chain in chondrocytes

indeed is β1, chondrocyte lysates were immunoprecipitated with antibodies against α10 orb β1 followed by Western blot using antibodies against the β1-subunit (Figure 8b). These results clearly demonstrated that α10 is a member of the β1-integrin family. However, the results do not exclude the possibility that α10 can associate with other β-chains in other situations.

Example 8

Immunohistochemical staining of the integrin subunit α10 in human and mouse cartilage

Material and methods

Frozen sections of adult cartilage (trochlear groove) obtained during surgery (provided by Anders Lindahl, Salgrenska Hospital, Gothenburg, Sweden and frozen sections from of 3 day old mouse limb were fixed and prepared for immunohistochemistry as earlier describ-ed (Camper et al, JBC, 273, 20383-20389 (1998)). Expres-sion of α10 integrin subunit was analysed using the poly-clonal antibody against the cytoplasmic domain as a pri-mary antibody (see Example 6) and a secondary antibody conjugated to peroxidase.

Results

Figures 9 show immunostaining of human adult articu-lar cartilage.

The α10-antibody recognising the cytoplasmic domain of α10 stained the chondrocytes in tissue sections of human articular cartilage (A). The staining was depleted when the antibody was preincubated with the α10- peptide (B). A control antibody recognising the α9 integrin sub-unit did not bind to the chondrocyte (C).

Figures 10 shows that the α10 antibody stain the majority of chondrocytes in the growing bone anlage (a and b). The α10 antibody also recognised cells in the ossification groove of Ranvier (b), especially the osteoblast in the bone bark which are lining the carti-lage in the metaphys are highly positive for α10. The

cells in the ossification groove of Ranvier are believed to be important for the growth in diameter of the bone. The integrin subunit α10 is also highly expressed in perichondrium and periosteum. Cell in these tissues are likely important in the repair of the cartilage tissue. The described localisation of the integrin subunit α10 suggest that this integrin is important for the function of the cartilage tissue.

Example 9

Immunohistochemical staining of the integrin subunit α10 during mouse development

Material and methods

Frozen sections from mouse embryos (13.5 days) were investigated for expression of α10 by immunhistochemistry as described in Camper et al, JBC, 273, 20383-20389 (1998). Expression of α10 integrin subunit was analysed using the polyclonal antibody against the cytoplasmic domain as a primary antibody (see Example 6) and a secondary antibody conjugated to peroxidase. The embryo sections were also investigated for expression of integrin subunit α1 (monoclonal antibody from Pharmingen) and collagen type II (monoclonal antibody, kind gift from Dr John Mo, Lund University, Sweden).

Results

Figure 11 show that α10 integrin subunit is unregulated in the limb when the mesenchymal cells undergo condensation to form cartilage (a). Especially the edge of the newly formed cartilage has high expression of α10. The formation of cartilage is verified by the high expression of the cartilage specific collage type II (b). The control antibody against α1 integrin subunit showed only weak expression on the cartilage (c). In other experiments expression of α10 was found in all cartilage containing tissues in the 3 day old mouse including limbs, ribs and vertebrae. The upregulation of α10 during formation of cartilage suggest that this integrin subunit is

important both in the development of cartilage and bone and in the repair of damaged cartilage tissue.

Example 10

mRNA expression of α10 in tissues other than articular cartilage

Material and methods

Expression of α10 integrin subunit was examined on the mRNA level in different human tissues. A Northern dot blot with immobilised mRNA from the listed tissues in Figure 12 was hybridised with an α10 integrin cDNA probe isolated from the race 1-containing plasmid using the restriction enzymes *Bam*H1 and *Nco*1. The degree of hybridisation was analysed using a phospho imager. The following symbols denote mRNA level in increasing order: -, +, ++, +++, ++++.

Results

Analysis of the hybridised mRNA showed that α10 was expressed in aorta, trachea, spinal cord, heart, lung, and kidney (Figure 12). All other tissues appeared negative for α10 expression. These results point to a restricted distribution of the α10 integrin subunit.

Example 11

Immunohistochemical staining of α10 in fascia around tendon and skeletal muscle and in tendon structures in heart valves.

Materials and methods

Frozen sections of adult cartilage (trochlear groove) obtained during surgery (provided by Anders Lindahl, Salgrenska Hospital, Gothenburg, Sweden and frozen sections from of 3 day old mouse limb were fixed and prepared for immunohistochemistry as earlier described (Camper et al, JBC, 273, 20383-20389 (1998)). Expression of α10 integrin subunit was analysed using the polyclonal antibody against the cytoplasmic domain as a pri-

mary antibody (see Example 6) and a secondary antibody conjugated to peroxidase.

Results

As shown in figures 13 expression of α10 was found in the fascia surrounding tendon (a) and skeletal muscle (b) and in the tendon structures in the heart valves (c). This localisation suggest that α10 can bind to other matrix molecules in addition to the cartilage specific collagen type II. The localisation of the integrin α10 on the surface of tendons indicate that α10 can be involved in unwanted adhesion that often occurs between tendon/ ligaments and the surrounding tissue after infection, injury or after surgery.

Example 12

mRNA expression of α10 integrin subuhit in chondrocytes, endothelial cells and fibroblasts.

Material and methods

Isolation of mRNA, synthesis of cDNA and PCR amplification was done as earlier described (Camper et al, JBC, 273, 20383-20389 (1998)).

Results

Figure 14 shows PCR amplification of α10 cDNA from human articular chondrocytes (lanes A6 and B1), human umbilical vein endothelial cells (lane A2), human fibroblasts (lane A4) and rat tendon (Fig 14b, lane B2). Lanes 1, 3, and 5 in figure 14 A show amplified fragments corresponding to the integrin subunit α2 in endothelial cells, fibroblasts and chondrocytes, respectively. cDNA-primers corresponding to the α10 sequence positions nt 2919-2943 (forward) and nt 3554-3578 (reverse) (see Figure 6) were used to amplify α10 cDNA from the different cells. The figure shows that α10 was amplified in all three cell types. Two fragments of α10 was amplified which represent the intact form of α10 (larger fragment) and a splice variant (smaller fragment). The larger frag-

ment was dominating in chondrocytes while the smaller fragment was more pronounced in tendon (B2).

Example 13

Construction of α10 mammalian expression vector.

The full length protein coding sequence of α10 (combined from 3 clones, see figure 6) was inserted into the mammalian expression vector, pcDNA3.1/Zeo (Invitrogen). The vector contains SV40 promoter and Zeosin selection sequence. The α10 containing expression vector was transfected into cells that express the β1-integrin subunit but lack expression of the α10 subunit. Expression of the α10 integrin subunit on the cell surface can be analysed by immunoprecipitation and/or flow cytometry using antibodies specific for α10. The ligand binding capacity and the function of the inserted α10 integrin subunit can be demonstrated in cell adhesion experiment and in signalling experiments.

Example 14

Construction of mammalian expression vector containing a splice variant of α10.

The full length protein coding sequence of the splice variant of α10 (nt 2942-nt3055 deleted) was inserted into the mammalian expression vector pcDNA3 (see Example 13). Expression and function of the splice variant can be analysed as described in example 13 and compared with the intact α10 integrin subunit.

Example 15

Partial isolation and characterisation of the α10 integrin genomic DNA

Material and methods

Human α10 cDNA, isolated from the racel-containing plasmid using the restriction enzymes *Bam*HI (GIBCO BRL) and *Nco*I (Boehringer Mannheim), was ^{32}P-labelled and used as a probe for screening of a mouse 129 cosmid library

(provided by Reinhard Fässler, Lund University). Positive clones were isolated and subcloned. Selected plasmids were purified and sequenced as described earlier (Camper et al, JBC, 273, 20383-20389 (1998)) using T3, T7 and internal specific primers. Primers corresponding to mouse genomic DNA were then constructed and used in PCR to amplify and identify the genomic sequence of α10 from the cosmid clones.

Results

Figure 15 shows 7958 nt of the α10 gene. This partial genomic DNA sequence of α10 integrin contains 8 exons, and a Kozak sequence. The mouse genomic α10 sequence was used to generate a targeting vector for knockout experiments.

Example 16

Upregulation of α10 integrin subunit in chondrocytes cultured in alginate beads

Material and methods

Human chondrocytes cultured in monolayer for 2 weeks were detached with trypsin-EDTA and introduced into alginate beads. Chondrocytes cultured in alginate are known to preserve their phenotype while chondrocytes cultured in monolayer are dedifferentiated. After 11 days chondrocytes cultured either in alginate or on monolayer were isolated and surface labelled with ^{125}I. The α10 integrin subunit was then immunoprecipitated with polyclonal antibodies recognising the cytoplasmic domain of α10 (see Example 6 and Camper et al, JBC, 273, 20383-20389 (1998)).

Results

As shown in figure 16 chondrocytes cultured in alginate beads (lanes 3 and 4) upregulated their protein expression of α10β1. This was in contrast to chondrocytes cultured in monolayer (lanes 1 and 2) which had a very low expression of α10β1. Immunoprecipitation with ab control antibody is shown in lanes 1 and 3.It is known that

chondrocytes preserve their cartilage specific matrixproduction in alginate cultures but not in monolayer culture which point to that alginate preserve the phenotype of chondrocytes. These results support that α10 integrin subunit can be used as a marker for differentiated chondrocytes.

Example 17

Immunoprecipitation of the α10 integrin subunit from human smooth muscle cells.

Material and methods

Human smooth muscle cells were isolated from human aorta. After one week in culture the cells were ^{125}I-labelled, lysed and immunoprecipitated with antibodies against the integrin subunit β1 (lane 1), α1 (lane 2), α2 (lane 3), α10 (lane 4), α3 (lane 5), control (lane 6) (Figure 17). The experiment was done as described in Example 7.

Results

The α10 antibody precipitated two bands from the smooth muscle cells corresponding to the α10 and the β1 integrin subunit (Fig. 17).

Example 18

Construction of bacterial expression vector containing sequence for α10 splice region.

A plasmid for intracellular expression in E. coli of the alternatively spliced region (amino acid pos. 952-986, SEQ. ID 1) was constructed as described. The alternatively spliced region were back-translated using the E. coli high frequency codon table, creating a cDNA sequence of 96% identity with the original sequence (SEQ. ID 1 nucleotide pos 2940-3044). Using sequence overlap extension (Horton et al., Biotechniques 8:528, 1990) primer α10pfor (tab. I) and α10prev (tab. I) was used to generate a double stranded fragment encoding the α10 amino acid sequence. This fragment was used as a PCR

template with primers α10pfor2 (tab. I) and α10prev2 (tab. I) in order to generate restriction enzyme site for sub-cloning in a pET vector containing the Z-domain of staphylococcal protein A, creating a fusion of the α10 spliced region with the amino terminal of the Z-domain with trombin cleavage site residing in-between. The fragment generated in the second PCR reaction is shown (SEQ ID No. 3) also indicating the unique restriction enzymes used for sub-cloning in the expression vector.

Table I

α10pfor	5'-GTTCAGAACCTGGGTTGCTACGTTGTTTCCGGTCTGATCATCTCCGCTCTGCTGCCGGCTGT-3'
α10pfor2	5'-GGGGCATATGGTTCAGAACCTGGGTTGCTACGTTG-3'
α10prev	5'-GATAACCTGGGACAAGCTTAGGAAGTAGTTACCACCGTGAGCAACAGCCGGCAGCAGAGCGGA-3'
α10prev2	5'-GGGGGGATCCGCGCGGCACCAGGCCGCTGATAACCTGGGACAAGCTTAGGAAGT-3'

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SEQUENCE LISTING

(1) GENERAL INFORMATION:
  (i) NUMBER OF SEQUENCES: 2

(2) INFORMATION FOR SEQ ID NO. 1:
  (i) SEQUENCE CHARACTERISTICS:
    (A) LENGTH: 3884 base pairs
    (B) TYPE: nucleic acid and amino acid
    (C) STRANDEDNESS: double
    (D) TOPOLOGY: linear

  (ii) MOLECULAR TYPE: cDNA
  (vi) ORIGINAL SOURCE:
    (E) ORGANISM: human
    (F) CELLTYPE: chondrocyte
  (xi) SEQUENCE DESCRIPTION: SEQ ID NO. 1:

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        CAGGTCAGAAACCGATCAGGCATGGAACTCCCCTTCGTCACTCACCTGTTCTTGCCCCTG
      1 ---------+---------+---------+---------+---------+---------+ 60
        GTCCAGTCTTTGGCTAGTCCGTACCTTGAGGGGAAGCAGTGAGTGGACAAGAACGGGGAC

a                               M  E  L  P  F  V  T  H  L  F  L  P  L  -

        GTGTTCCTGACAGGTCTCTGCTCCCCCTTTAACCTGGATGAACATCACCCACGCCTATTC
     61 ---------+---------+---------+---------+---------+---------+ 120
        CACAAGGACTGTCCAGAGACGAGGGGGAAATTGGACCTACTTGTAGTGGGTGCGGATAAG

a         V  F  L  T  G  L  C  S  P  F  N  L  D  E  H  H  P  R  L  F  -

        CCAGGGCCACCAGAAGCTGAATTTGGATACAGTGTCTTACAACATGTTGGGGGTGGACAG
    121 ---------+---------+---------+---------+---------+---------+ 180
        GGTCCCGGTGGTCTTCGACTTAAACCTATGTCACAGAATGTTGTACAACCCCCACCTGTC

a         P  G  P  P  E  A  E  F  G  Y  S  V  L  Q  H  V  G  G  G  Q  -

        CGATGGATGCTGGTGGGCGCCCCCTGGGATGGGCCTTCAGGCGACCGGAGGGGGGACGTT
    181 ---------+---------+---------+---------+---------+---------+ 240
        GCTACCTACGACCACCCGCGGGGGACCCTACCCGGAAGTCCGCTGGCCTCCCCCCTGCAA

a         R  W  M  L  V  G  A  P  W  D  G  P  S  G  D  R  R  G  D  V  -

        TATCGCTGCCCTGTAGGGGGGGCCCACAATGCCCCATGTGCCAAGGGCCACTTAGGTGAC
    241 ---------+---------+---------+---------+---------+---------+ 300
        ATAGCGACGGGACATCCCCCCCGGGTGTTACGGGGTACACGGTTCCCGGTGAATCCACTG

a         Y  R  C  P  V  G  G  A  H  N  A  P  C  A  K  G  H  L  G  D  -

        TACCAACTGGGAAATTCATCTCATCCTGCTGTGAATATGCACCTGGGGATGTCTCTGTTA
    301 ---------+---------+---------+---------+---------+---------+ 360
        ATGGTTGACCCTTTAAGTAGAGTAGGACGACACTTATACGTGGACCCCTACAGAGACAAT

a         Y  Q  L  G  N  S  S  H  P  A  V  N  M  H  L  G  M  S  L  L  -

        GAGACAGATGGTGATGGGGGATTCATGGCCTGTGCCCCTCTCTGGTCTCGTGCTTGTGGC
    361 ---------+---------+---------+---------+---------+---------+ 420
        CTCTGTCTACCACTACCCCCTAAGTACCGGACACGGGGAGAGACCAGAGCACGAACACCG

a         E  T  D  G  D  G  G  F  M  A  C  A  P  L  W  S  R  A  C  G  -
```

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        AGCTCTGTCTTCAGTTCTGGGATATGTGCCCGTGTGGATGCTTCATTCCAGCCTCAGGGA
     421 ---------+---------+---------+---------+---------+---------+ 480
        TCGAGACAGAAGTCAAGACCCTATACACGGGCACACCTACGAAGTAAGGTCGGAGTCCCT

a       S  S  V  F  S  S  G  I  C  A  R  V  D  A  S  F  Q  P  Q  G  -

        AGCCTGGCACCCACTGCCCAACGCTGCCCAACATACATGGATGTTGTCATTGTCTTGGAT
     481 ---------+---------+---------+---------+---------+---------+ 540
        TCGGACCGTGGGTGACGGGTTGCGACGGGTTGTATGTACCTACAACAGTAACAGAACCTA

a       S  L  A  P  T  A  Q  R  C  P  T  Y  M  D  V  V  I  V  L  D  -

        GGCTCCAACAGCATCTACCCCTGGTCTGAAGTTCAGACCTTCCTACGAAGACTGGTAGGG
     541 ---------+---------+---------+---------+---------+---------+ 600
        CCGAGGTTGTCGTAGATGGGGACCAGACTTCAAGTCTGGAAGGATGCTTCTGACCATCCC

a       G  S  N  S  I  Y  P  W  S  E  V  Q  T  F  L  R  R  L  V  G  -

        AAACTGTTTATTGACCCAGAACAGATACAGGTGGGACTGGTACAGTATGGGGAGAGCCCT
     601 ---------+---------+---------+---------+---------+---------+ 660
        TTTGACAAATAACTGGGTCTTGTCTATGTCCACCCTGACCATGTCATACCCCTCTCGGGA

a       K  L  F  I  D  P  E  Q  I  Q  V  G  L  V  Q  Y  G  E  S  P  -

        GTACATGAGTGGTCCCTGGGAGATTTCCGAACGAAGGAAGAAGTGGTGAGAGCAGCAAAG
     661 ---------+---------+---------+---------+---------+---------+ 720
        CATGTACTCACCAGGGACCCTCTAAAGGCTTGCTTCCTTCTTCACCACTCTCGTCGTTTC

a       V  H  E  W  S  L  G  D  F  R  T  K  E  E  V  V  R  A  A  K  -

        AACCTCAGTCGGCGGGAGGGACGAGAAACAAAGACTGCCCAAGCAATAATGGTGGCCTGC
     721 ---------+---------+---------+---------+---------+---------+ 780
        TTGGAGTCAGCCGCCCTCCCTGCTCTTTGTTTCTGACGGGTTCGTTATTACCACCGGACG

a       N  L  S  R  R  E  G  R  E  T  K  T  A  Q  A  I  M  V  A  C  -

        ACAGAAGGGTTCAGTCAGTCCCATGGGGGCCGACCCGAGGCTGCCAGGCTACTGGTGGTT
     781 ---------+---------+---------+---------+---------+---------+ 840
        TGTCTTCCCAAGTCAGTCAGGGTACCCCCGGCTGGGCTCCGACGGTCCGATGACCACCAA

a       T  E  G  F  S  Q  S  H  G  G  R  P  E  A  A  R  L  L  V  V  -

        GTCACTGATGGAGAGTCCCATGATGGAGAGGAGCTTCCTGCAGCACTAAAGGCCTGTGAG
     841 ---------+---------+---------+---------+---------+---------+ 900
        CAGTGACTACCTCTCAGGGTACTACCTCTCCTCGAAGGACGTCGTGATTTCCGGACACTC

a       V  T  D  G  E  S  H  D  G  E  E  L  P  A  A  L  K  A  C  E  -

        GCTGGAAGAGTGACACGCTATGGGATTGCAGTCCTTGGTCACTACCTCCGGCGGCAGCGA
     901 ---------+---------+---------+---------+---------+---------+ 960
        CGACCTTCTCACTGTGCGATACCCTAACGTCAGGAACCAGTGATGGAGGCCGCCGTCGCT

a       A  G  R  V  T  R  Y  G  I  A  V  L  G  H  Y  L  R  R  Q  R  -

        GATCCCAGCTCTTTCCTGAGAGAAATTAGAACTATTGCCAGTGATCCAGATGAGCGATTC
     961 ---------+---------+---------+---------+---------+---------+ 1020
        CTAGGGTCGAGAAAGGACTCTCTTTAATCTTGATAACGGTCACTAGGTCTACTCGCTAAG

a       D  P  S  S  F  L  R  E  I  R  T  I  A  S  D  P  D  E  R  F  -

        TTCTTCAATGTCACAGATGAGGCTGCTCTGACTGACATTGTGGATGCACTAGGAGATCGG
    1021 ---------+---------+---------+---------+---------+---------+ 1080
        AAGAAGTTACAGTGTCTACTCCGACGAGACTGACTGTAACACCTACGTGATCCTCTAGCC

a       F  F  N  V  T  D  E  A  A  L  T  D  I  V  D  A  L  G  D  R  -
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          ATTTTTGGCCTTGAAGGGTCCCATGCAGAAAACGAAAGCTCCTTTGGGCTGGAAATGTCT
     1081 ---------+---------+---------+---------+---------+---------+ 1140
          TAAAAACCGGAACTTCCCAGGGTACGTCTTTTGCTTTCGAGGAAACCCGACCTTTACAGA

a          I  F  G  L  E  G  S  H  A  E  N  E  S  S  F  G  L  E  M  S  -

          CAGATTGGTTTCTCCACTCATCGGCTAAAGGATGGGATTCTTTTTGGGATGGTGGGGGCC
     1141 ---------+---------+---------+---------+---------+---------+ 1200
          GTCTAACCAAAGAGGTGAGTAGCCGATTTCCTACCCTAAGAAAAACCCTACCACCCCCGG

a          Q  I  G  F  S  T  H  R  L  K  D  G  I  L  F  G  M  V  G  A  -

          TATGACTGGGGAGGCTCTGTGCTATGGCTTGAAGGAGGCCACCGCCTTTTCCCCCCACGA
     1201 ---------+---------+---------+---------+---------+---------+ 1260
          ATACTGACCCCTCCGAGACACGATACCGAACTTCCTCCGGTGGCGGAAAAGGGGGGTGCT

a          Y  D  W  G  G  S  V  L  W  L  E  G  G  H  R  L  F  P  P  R  -

          ATGGCACTGGAAGACGAGTTCCCCCCTGCACTGCAGAACCATGCAGCCTACCTGGGTTAC
     1261 ---------+---------+---------+---------+---------+---------+ 1320
          TACCGTGACCTTCTGCTCAAGGGGGGACGTGACGTCTTGGTACGTCGGATGGACCCAATG

a          M  A  L  E  D  E  F  P  P  A  L  Q  N  H  A  A  Y  L  G  Y  -

          TCTGTTTCTTCCATGCTTTTGCGGGGTGGACGCCGCCTGTTTCTCTCTGGGGCTCCTCGA
     1321 ---------+---------+---------+---------+---------+---------+ 1380
          AGACAAAGAAGGTACGAAAACGCCCCACCTGCGGCGGACAAAGAGAGACCCCGAGGAGCT

a          S  V  S  S  M  L  L  R  G  G  R  R  L  F  L  S  G  A  P  R  -

          TTTAGACATCGAGGAAAAGTCATCGCCTTCCAGCTTAAGAAAGATGGGGCTGTGAGGGTT
     1381 ---------+---------+---------+---------+---------+---------+ 1440
          AAATCTGTAGCTCCTTTTCAGTAGCGGAAGGTCGAATTCTTTCTACCCCGACACTCCCAA

a          F  R  H  R  G  K  V  I  A  F  Q  L  K  K  D  G  A  V  R  V  -

          GCCCAGAGCCTCCAGGGGGAGCAGATTGGTTCATACTTTGGCAGTGAGCTCTGCCCATTG
     1441 ---------+---------+---------+---------+---------+---------+ 1500
          CGGGTCTCGGAGGTCCCCCTCGTCTAACCAAGTATGAAACCGTCACTCGAGACGGGTAAC

a          A  Q  S  L  Q  G  E  Q  I  G  S  Y  F  G  S  E  L  C  P  L  -

          GATACAGATAGGGATGGAACAACTGATGTCTTACTTGTGGCTGCCCCCATGTTCCTGGGA
     1501 ---------+---------+---------+---------+---------+---------+ 1560
          CTATGTCTATCCCTACCTTGTTGACTACAGAATGAACACCGACGGGGGTACAAGGACCCT

a          D  T  D  R  D  G  T  T  D  V  L  L  V  A  A  P  M  F  L  G  -

          CCCCAGAACAAGGAAACAGGACGTGTTTATGTGTATCTGGTAGGCCAGCAGTCCTTGCTG
     1561 ---------+---------+---------+---------+---------+---------+ 1620
          GGGGTCTTGTTCCTTTGTCCTGCACAAATACACATAGACCATCCGGTCGTCAGGAACGAC

a          P  Q  N  K  E  T  G  R  V  Y  V  Y  L  V  G  Q  Q  S  L  L  -

          ACCCTCCAAGGAACACTTCAGCCAGAACCCCCCCAGGATGCTCGGTTTGGCTTTGCCATG
     1621 ---------+---------+---------+---------+---------+---------+ 1680
          TGGGAGGTTCCTTGTGAAGTCGGTCTTGGGGGGGTCCTACGAGCCAAACCGAAACGGTAC

a          T  L  Q  G  T  L  Q  P  E  P  P  Q  D  A  R  F  G  F  A  M  -

          GGAGCTCTTCCTGATCTGAACCAAGATGGTTTTGCTGATGTGGCTGTGGGGGCGCCTCTG
     1681 ---------+---------+---------+---------+---------+---------+ 1740
          CCTCGAGAAGGACTAGACTTGGTTCTACCAAAACGACTACACCGACACCCCCGCGGAGAC

a          G  A  L  P  D  L  N  Q  D  G  F  A  D  V  A  V  G  A  P  L  -
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```
            GAAGATGGGCACCAGGGAGCACTGTACCTGTACCATGGAACCCAGAGTGGAGTCAGGCCC
      1741  ---------+---------+---------+---------+---------+---------+ 1800
            CTTCTACCCGTGGTCCCTCGTGACATGGACATGGTACCTTGGGTCTCACCTCAGTCCGGG

a           E  D  G  H  Q  G  A  L  Y  L  Y  H  G  T  Q  S  G  V  R  P  -

            CATCCTGCCCAGAGGATTGCTGCTGCCTCCATGCCACATGCCCTCAGCTACTTTGGCCGA
      1801  ---------+---------+---------+---------+---------+---------+ 1860
            GTAGGACGGGTCTCCTAACGACGACGGAGGTACGGTGTACGGGAGTCGATGAAACCGGCT

a           H  P  A  Q  R  I  A  A  A  S  M  P  H  A  L  S  Y  F  G  R  -

            AGTGTGGATGGTCGGCTAGATCTGGATGGAGATGATCTGGTCGATGTGGCTGTGGGTGCC
      1861  ---------+---------+---------+---------+---------+---------+ 1920
            TCACACCTACCAGCCGATCTAGACCTACCTCTACTAGACCAGCTACACCGACACCCACGG

a           S  V  D  G  R  L  D  L  D  G  D  D  L  V  D  V  A  V  G  A  -

            CAGGGGGCAGCCATCCTGCTCAGCTCCCGGCCCATTGTCCATCTGACCCCATCACTGGAG
      1921  ---------+---------+---------+---------+---------+---------+ 1980
            GTCCCCCGTCGGTAGGACGAGTCGAGGGCCGGGTAACAGGTAGACTGGGGTAGTGACCTC

a           Q  G  A  A  I  L  L  S  S  R  P  I  V  H  L  T  P  S  L  E  -

            GTGACCCCACAGGCCATCAGTGTGGTTCAGAGGGACTGTAGGCGGCGAGGCCAAGAAGCA
      1981  ---------+---------+---------+---------+---------+---------+ 2040
            CACTGGGGTGTCCGGTAGTCACACCAAGTCTCCCTGACATCCGCCGCTCCGGTTCTTCGT

a           V  T  P  Q  A  I  S  V  V  Q  R  D  C  R  R  R  G  Q  E  A  -

            GTCTGTCTGACTGCAGCCCTTTGCTTCCAAGTGACCTCCCGTACTCCTGGTCGCTGGGAT
      2041  ---------+---------+---------+---------+---------+---------+ 2100
            CAGACAGACTGACGTCGGGAAACGAAGGTTCACTGGAGGGCATGAGGACCAGCGACCCTA

a           V  C  L  T  A  A  L  C  F  Q  V  T  S  R  T  P  G  R  W  D  -

            CACCAATTCTACATGAGGTTCACCGCATCACTGGATGAATGGACTGCTGGGGCACGTGCA
      2101  ---------+---------+---------+---------+---------+---------+ 2160
            GTGGTTAAGATGTACTCCAAGTGGCGTAGTGACCTACTTACCTGACGACCCCGTGCACGT

a           H  Q  F  Y  M  R  F  T  A  S  L  D  E  W  T  A  G  A  R  A  -

            GCATTTGATGGCTCTGGCCAGAGGTTGTCCCCTCGGAGGCTCCGGCTCAGTGTGGGGAAT
      2161  ---------+---------+---------+---------+---------+---------+ 2220
            CGTAAACTACCGAGACCGGTCTCCAACAGGGGAGCCTCCGAGGCCGAGTCACACCCCTTA

a           A  F  D  G  S  G  Q  R  L  S  P  R  R  L  R  L  S  V  G  N  -

            GTCACTTGTGAGCAGCTACACTTCCATGTGCTGGATACATCAGATTACCTCCGGCCAGTG
      2221  ---------+---------+---------+---------+---------+---------+ 2280
            CAGTGAACACTCGTCGATGTGAAGGTACACGACCTATGTAGTCTAATGGAGGCCGGTCAC

a           V  T  C  E  Q  L  H  F  H  V  L  D  T  S  D  Y  L  R  P  V  -

            GCCTTGACTGTGACCTTTGCCTTGGACAATACTACAAAGCCAGGGCCTGTGCTGAATGAG
      2281  ---------+---------+---------+---------+---------+---------+ 2340
            CGGAACTGACACTGGAAACGGAACCTGTTATGATGTTTCGGTCCCGGACACGACTTACTC

a           A  L  T  V  T  F  A  L  D  N  T  T  K  P  G  P  V  L  N  E  -

            GGCTCACCCACCTCTATACAAAAGCTGGTCCCCTTCTCAAAGGATTGTGGCCCTGACAAT
      2341  ---------+---------+---------+---------+---------+---------+ 2400
            CCGAGTGGGTGGAGATATGTTTTCGACCAGGGGAAGAGTTTCCTAACACCGGGACTGTTA

a           G  S  P  T  S  I  Q  K  L  V  P  F  S  K  D  C  G  P  D  N  -
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```
      GAATGTGTCACAGACCTGGTGCTTCAAGTGAATATGGACATCAGAGGCTCCAGGAAGGCC
 2401 ---------+---------+---------+---------+---------+---------+ 2460
      CTTACACAGTGTCTGGACCACGAAGTTCACTTATACCTGTAGTCTCCGAGGTCCTTCCGG

a     E  C  V  T  D  L  V  L  Q  V  N  M  D  I  R  G  S  R  K  A  -

      CCATTTGTGGTTCGAGGTGGCCGGCGGAAAGTGCTGGTATCTACAACTCTGGAGAACAGA
 2461 ---------+---------+---------+---------+---------+---------+ 2520
      GGTAAACACCAAGCTCCACCGGCCGCCTTTCACGACCATAGATGTTGAGACCTCTTGTCT

a     P  F  V  V  R  G  G  R  R  K  V  L  V  S  T  T  L  E  N  R  -

      AAGGAAAATGCTTACAATACGAGCCTGAGTATCATCTTCTCTAGAAACCTCCACCTGGCC
 2521 ---------+---------+---------+---------+---------+---------+ 2580
      TTCCTTTTACGAATGTTATGCTCGGACTCATAGTAGAAGAGATCTTTGGAGGTGGACCGG

a     K  E  N  A  Y  N  T  S  L  S  I  I  F  S  R  N  L  H  L  A  -

      AGTCTCACTCCTCAGAGAGAGAGCCCAATAAAGGTGGAATGTGCCGCCCCTTCTGCTCAT
 2581 ---------+---------+---------+---------+---------+---------+ 2640
      TCAGAGTGAGGAGTCTCTCTCTCGGGTTATTTCCACCTTACACGGCGGGGAAGACGAGTA

a     S  L  T  P  Q  R  E  S  P  I  K  V  E  C  A  A  P  S  A  H  -

      GCCCGGCTCTGCAGTGTGGGGCATCCTGTCTTCCAGACTGGAGCCAAGGTGACCTTTCTG
 2641 ---------+---------+---------+---------+---------+---------+ 2700
      CGGGCCGAGACGTCACACCCCGTAGGACAGAAGGTCTGACCTCGGTTCCACTGGAAAGAC

a     A  R  L  C  S  V  G  H  P  V  F  Q  T  G  A  K  V  T  F  L  -

      CTAGAGTTTGAGTTTAGCTGCTCCTCTCTCCTGAGCCAGGTCTTTGGGAAGCTGACTGCC
 2701 ---------+---------+---------+---------+---------+---------+ 2760
      GATCTCAAACTCAAATCGACGAGGAGAGAGGACTCGGTCCAGAAACCCTTCGACTGACGG

a     L  E  F  E  F  S  C  S  S  L  L  S  Q  V  F  G  K  L  T  A  -

      AGCAGTGACAGCCTGGAGAGAAATGGCACCCTTCAAGAAAACACAGCCCAGACCTCAGCC
 2761 ---------+---------+---------+---------+---------+---------+ 2820
      TCGTCACTGTCGGACCTCTCTTTACCGTGGGAAGTTCTTTTGTGTCGGGTCTGGAGTCGG

a     S  S  D  S  L  E  R  N  G  T  L  Q  E  N  T  A  Q  T  S  A  -

      TACATCCAATATGAGCCCCACCTCCTGTTCTCTAGTGAGTCTACCCTGCACCGCTATGAG
 2821 ---------+---------+---------+---------+---------+---------+ 2880
      ATGTAGGTTATACTCGGGGTGGAGGACAAGAGATCACTCAGATGGGACGTGGCGATACTC

a     Y  I  Q  Y  E  P  H  L  L  F  S  S  E  S  T  L  H  R  Y  E  -

      GTTCACCCATATGGGACCCTCCCAGTGGGTCCTGGCCCAGAATTCAAAACCACTCTCAGG
 2881 ---------+---------+---------+---------+---------+---------+ 2940
      CAAGTGGGTATACCCTGGGAGGGTCACCCAGGACCGGGTCTTAAGTTTTGGTGAGAGTCC

a     V  H  P  Y  G  T  L  P  V  G  P  G  P  E  F  K  T  T  L  R  -

      GTTCAGAACCTAGGCTGCTATGTGGTCAGTGGCCTCATCATCTCAGCCCTCCTTCCAGCT
 2941 ---------+---------+---------+---------+---------+---------+ 3000
      CAAGTCTTGGATCCGACGATACACCAGTCACCGGAGTAGTAGAGTCGGGAGGAAGGTCGA

a     V  Q  N  L  G  C  Y  V  V  S  G  L  I  I  S  A  L  L  P  A  -

      GTGGCCCATGGGGGCAATTACTTCCTATCACTGTCTCAAGTCATCACTAACAATGCAAGC
 3001 ---------+---------+---------+---------+---------+---------+ 3060
      CACCGGGTACCCCCGTTAATGAAGGATAGTGACAGAGTTCAGTAGTGATTGTTACGTTCG

a     V  A  H  G  G  N  Y  F  L  S  L  S  Q  V  I  T  N  N  A  S  -
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     TGCATAGTGCAGAACCTGACTGAACCCCCAGGCCCACCTGTGCATCCAGAGGAGCTTCAA
3061 ---------+---------+---------+---------+---------+---------+ 3120
     ACGTATCACGTCTTGGACTGACTTGGGGGTCCGGGTGGACACGTAGGTCTCCTCGAAGTT

a    C  I  V  Q  N  L  T  E  P  P  G  P  P  V  H  P  E  E  L  Q  -

     CACACAAACAGACTGAATGGGAGCAATACTCAGTGTCAGGTGGTGAGGTGCCACCTTGGG
3121 ---------+---------+---------+---------+---------+---------+ 3180
     GTGTGTTTGTCTGACTTACCCTCGTTATGAGTCACAGTCCACCACTCCACGGTGGAACCC

a    H  T  N  R  L  N  G  S  N  T  Q  C  Q  V  V  R  C  H  L  G  -

     CAGCTGGCAAAGGGGACTGAGGTCTCTGTTGGACTATTGAGGCTGGTTCACAATGAATTT
3181 ---------+---------+---------+---------+---------+---------+ 3240
     GTCGACCGTTTCCCCTGACTCCAGAGACAACCTGATAACTCCGACCAAGTGTTACTTAAA

a    Q  L  A  K  G  T  E  V  S  V  G  L  L  R  L  V  H  N  E  F  -

     TTCCGAAGAGCCAAGTTCAAGTCCCTGACGGTGGTCAGCACCTTTGAGCTGGGAACCGAA
3241 ---------+---------+---------+---------+---------+---------+ 3300
     AAGGCTTCTCGGTTCAAGTTCAGGGACTGCCACCAGTCGTGGAAACTCGACCCTTGGCTT

a    F  R  R  A  K  F  K  S  L  T  V  V  S  T  F  E  L  G  T  E  -

     GAGGGCAGTGTCCTACAGCTGACTGAAGCCTCCCGTTGGAGTGAGAGCCTCTTGGAGGTG
3301 ---------+---------+---------+---------+---------+---------+ 3360
     CTCCCGTCACAGGATGTCGACTGACTTCGGAGGGCAACCTCACTCTCGGAGAACCTCCAC

a    E  G  S  V  L  Q  L  T  E  A  S  R  W  S  E  S  L  L  E  V  -

     GTTCAGACCCGGCCTATCCTCATCTCCCTGTGGATCCTCATAGGCAGTGTCCTGGGAGGG
3361 ---------+---------+---------+---------+---------+---------+ 3420
     CAAGTCTGGGCCGGATAGGAGTAGAGGGACACCTAGGAGTATCCGTCACAGGACCCTCCC

a    V  Q  T  R  P  I  L  I  S  L  W  I  L  I  G  S  V  L  G  G  -

     TTGCTCCTGCTTGCTCTCCTTGTCTTCTGCCTGTGGAAGCTTGGCTTCTTTGCCCATAAG
3421 ---------+---------+---------+---------+---------+---------+ 3480
     AACGAGGACGAACGAGAGGAACAGAAGACGGACACCTTCGAACCGAAGAAACGGGTATTC

a    L  L  L  L  A  L  L  V  F  C  L  W  K  L  G  F  F  A  H  K  -

     AAAATCCCTGAGGAAGAAAAAAGAGAAGAGAAGTTGGAGCAATGAATGTAGAATAAGGGT
3481 ---------+---------+---------+---------+---------+---------+ 3540
     TTTTAGGGACTCCTTCTTTTTTCTCTTCTCTTCAACCTCGTTACTTACATCTTATTCCCA

a    K  I  P  E  E  E  K  R  E  E  K  L  E  Q

     CTAGAAAGTCCTCCCTGGCAGCTTTCTTCAAGAGACTTGCATAAAAGCAGAGGTTTGGGG
3541 ---------+---------+---------+---------+---------+---------+ 3600
     GATCTTTCAGGAGGGACCGTCGAAAGAAGTTCTCTGAACGTATTTTCGTCTCCAAACCCC

     GCTCAGATGGGACAAGAAGCCGCCTCTGGACTATCTCCCCAGACCAGCAGCCTGACTTGA
3601 ---------+---------+---------+---------+---------+---------+ 3660
     CGAGTCTACCCTGTTCTTCGGCGGAGACCTGATAGAGGGGTCTGGTCGTCGGACTGAACT

     CTTTTGAGTCCTAGGGATGCTGCTGGCTAGAGATGAGGCTTTACCTCAGACAAGAAGAGC
3661 ---------+---------+---------+---------+---------+---------+ 3720
     GAAAACTCAGGATCCCTACGACGACCGATCTCTACTCCGAAATGGAGTCTGTTCTTCTCG
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```
     TGGCACCAAAACTAGCCATGCTCCCACCCTCTGCTTCCCTCCTCCTCGTGATCCTGGTTC
3721 ---------+---------+---------+---------+---------+---------+ 3780
     ACCGTGGTTTTGATCGGTACGAGGGTGGGAGACGAAGGGAGGAGGAGCACTAGGACCAAG

     CATAGCCAACACTGGGGCTTTTGTTTGGGGTCCTTTTATCCCCAGGAATCAATAATTTTT
3781 ---------+---------+---------+---------+---------+---------+ 3840
     GTATCGGTTGTGACCCCGAAAACAAACCCCAGGAAAATAGGGGTCCTTAGTTATTAAAAA

     TTGCCTAGGAAAAAAAAAAAGCGGCCGCGAATTCGATATCAAGCT
3841 ---------+---------+---------+---------+---- 3884
     AACGGATCCTTTTTTTTTTTCGCCGGCGCTTAAGCTATAGTTCGA
```

(2) INFORMATION FOR SEQ ID NO. 2:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 3779 base pairs
(B) TYPE: nucleid acid and amino acid
(C) STRANDEDNESS: double
(D) TOPOLOGY: linear
(E)
(i) MOLECULAR TYPE: cDNA
(vi) ORIGINAL SOURCE:
(A) ORGANISM: human
(B) CELLTYPE: chondrocyte

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 2:

```
      CAGGTCAGAAACCGATCAGGCATGGAACTCCCCTTCGTCACTCACCTGTTCTTGCCCCTG
   1  ---------+---------+---------+---------+---------+---------+ 60
      GTCCAGTCTTTGGCTAGTCCGTACCTTGAGGGGAAGCAGTGAGTGGACAAGAACGGGGAC

                          M  E  L  P  F  V  T  H  L  F  L  P  L  -

      GTGTTCCTGACAGGTCTCTGCTCCCCCTTTAACCTGGATGAACATCACCCACGCCTATTC
  61  ---------+---------+---------+---------+---------+---------+ 120
      CACAAGGACTGTCCAGAGACGAGGGGGAAATTGGACCTACTTGTAGTGGGTGCGGATAAG

a     V  F  L  T  G  L  C  S  P  F  N  L  D  E  H  H  P  R  L  F  -

      CCAGGGCCACCAGAAGCTGAATTTGGATACAGTGTCTTACAACATGTTGGGGGTGGACAG
 121  ---------+---------+---------+---------+---------+---------+ 180
      GGTCCCGGTGGTCTTCGACTTAAACCTATGTCACAGAATGTTGTACAACCCCCACCTGTC

a     P  G  P  P  E  A  E  F  G  Y  S  V  L  Q  H  V  G  G  G  Q  -

      CGATGGATGCTGGTGGGCGCCCCCTGGGATGGGCCTTCAGGCGACCGGAGGGGGGACGTT
 181  ---------+---------+---------+---------+---------+---------+ 240
      GCTACCTACGACCACCCGCGGGGGACCCTACCCGGAAGTCCGCTGGCCTCCCCCCTGCAA

a     R  W  M  L  V  G  A  P  W  D  G  P  S  G  D  R  R  G  D  V  -

      TATCGCTGCCCTGTAGGGGGGGCCCACAATGCCCCATGTGCCAAGGGCCACTTAGGTGAC
 241  ---------+---------+---------+---------+---------+---------+ 300
      ATAGCGACGGGACATCCCCCCCGGGTGTTACGGGGTACACGGTTCCCGGTGAATCCACTG

a     Y  R  C  P  V  G  G  A  H  N  A  P  C  A  K  G  H  L  G  D  -

      TACCAACTGGGAAATTCATCTCATCCTGCTGTGAATATGCACCTGGGGATGTCTCTGTTA
 301  ---------+---------+---------+---------+---------+---------+ 360
      ATGGTTGACCCTTTAAGTAGAGTAGGACGACACTTATACGTGGACCCCTACAGAGACAAT

a     Y  Q  L  G  N  S  S  H  P  A  V  N  M  H  L  G  M  S  L  L  -

      GAGACAGATGGTGATGGGGGATTCATGGCCTGTGCCCCTCTCTGGTCTCGTGCTTGTGGC
 361  ---------+---------+---------+---------+---------+---------+ 420
      CTCTGTCTACCACTACCCCCTAAGTACCGGACACGGGGAGAGACCAGAGCACGAACACCG

a     E  T  D  G  D  G  G  F  M  A  C  A  P  L  W  S  R  A  C  G  -

      AGCTCTGTCTTCAGTTCTGGGATATGTGCCCGTGTGGATGCTTCATTCCAGCCTCAGGGA
 421  ---------+---------+---------+---------+---------+---------+ 480
      TCGAGACAGAAGTCAAGACCCTATACACGGGCACACCTACGAAGTAAGGTCGGAGTCCCT
```

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a      S  S  V  F  S  S  G  I  C  A  R  V  D  A  S  F  Q  P  Q  G   -

       AGCCTGGCACCCACTGCCCAACGCTGCCCAACATACATGGATGTTGTCATTGTCTTGGAT
   481 ---------+---------+---------+---------+---------+---------+ 540
       TCGGACCGTGGGTGACGGGTTGCGACGGGTTGTATGTACCTACAACAGTAACAGAACCTA

a      S  L  A  P  T  A  Q  R  C  P  T  Y  M  D  V  V  I  V  L  D   -

       GGCTCCAACAGCATCTACCCCTGGTCTGAAGTTCAGACCTTCCTACGAAGACTGGTAGGG
   541 ---------+---------+---------+---------+---------+---------+ 600
       CCGAGGTTGTCGTAGATGGGGACCAGACTTCAAGTCTGGAAGGATGCTTCTGACCATCCC

a      G  S  N  S  I  Y  P  W  S  E  V  Q  T  F  L  R  R  L  V  G   -

       AAACTGTTTATTGACCCAGAACAGATACAGGTGGGACTGGTACAGTATGGGGAGAGCCCT
   601 ---------+---------+---------+---------+---------+---------+ 660
       TTTGACAAATAACTGGGTCTTGTCTATGTCCACCCTGACCATGTCATACCCCTCTCGGGA

a      K  L  F  I  D  P  E  Q  I  Q  V  G  L  V  Q  Y  G  E  S  P   -

       GTACATGAGTGGTCCCTGGGAGATTTCCGAACGAAGGAAGAAGTGGTGAGAGCAGCAAAG
   661 ---------+---------+---------+---------+---------+---------+ 720
       CATGTACTCACCAGGGACCCTCTAAAGGCTTGCTTCCTTCTTCACCACTCTCGTCGTTTC

a      V  H  E  W  S  L  G  D  F  R  T  K  E  E  V  V  R  A  A  K   -

       AACCTCAGTCGGCGGGAGGGACGAGAAACAAAGACTGCCCAAGCAATAATGGTGGCCTGC
   721 ---------+---------+---------+---------+---------+---------+ 780
       TTGGAGTCAGCCGCCCTCCCTGCTCTTTGTTTCTGACGGGTTCGTTATTACCACCGGACG

a      N  L  S  R  R  E  G  R  E  T  K  T  A  Q  A  I  M  V  A  C   -

       ACAGAAGGGTTCAGTCAGTCCCATGGGGGCCGACCCGAGGCTGCCAGGCTACTGGTGGTT
   781 ---------+---------+---------+---------+---------+---------+ 840
       TGTCTTCCCAAGTCAGTCAGGGTACCCCCGGCTGGGCTCCGACGGTCCGATGACCACCAA

a      T  E  G  F  S  Q  S  H  G  G  R  P  E  A  A  R  L  L  V  V   -

       GTCACTGATGGAGAGTCCCATGATGGAGAGGAGCTTCCTGCAGCACTAAAGGCCTGTGAG
   841 ---------+---------+---------+---------+---------+---------+ 900
       CAGTGACTACCTCTCAGGGTACTACCTCTCCTCGAAGGACGTCGTGATTTCCGGACACTC

a      V  T  D  G  E  S  H  D  G  E  E  L  P  A  A  L  K  A  C  E   -

       GCTGGAAGAGTGACACGCTATGGGATTGCAGTCCTTGGTCACTACCTCCGGCGGCAGCGA
   901 ---------+---------+---------+---------+---------+---------+ 960
       CGACCTTCTCACTGTGCGATACCCTAACGTCAGGAACCAGTGATGGAGGCCGCCGTCGCT

a      A  G  R  V  T  R  Y  G  I  A  V  L  G  H  Y  L  R  R  Q  R   -

       GATCCCAGCTCTTTCCTGAGAGAAATTAGAACTATTGCCAGTGATCCAGATGAGCGATTC
   961 ---------+---------+---------+---------+---------+---------+ 1020
       CTAGGGTCGAGAAAGGACTCTCTTTAATCTTGATAACGGTCACTAGGTCTACTCGCTAAG

a      D  P  S  S  F  L  R  E  I  R  T  I  A  S  D  P  D  E  R  F   -

       TTCTTCAATGTCACAGATGAGGCTGCTCTGACTGACATTGTGGATGCACTAGGAGATCGG
  1021 ---------+---------+---------+---------+---------+---------+ 1080
       AAGAAGTTACAGTGTCTACTCCGACGAGACTGACTGTAACACCTACGTGATCCTCTAGCC

a      F  F  N  V  T  D  E  A  A  L  T  D  I  V  D  A  L  G  D  R   -

       ATTTTTGGCCTTGAAGGGTCCCATGCAGAAAACGAAAGCTCCTTTGGGCTGGAAATGTCT
  1081 ---------+---------+---------+---------+---------+---------+ 1140
       TAAAAACCGGAACTTCCCAGGGTACGTCTTTTGCTTTCGAGGAAACCCGACCTTTACAGA
```

```
a        I  F  G  L  E  G  S  H  A  E  N  E  S  S  F  G  L  E  M  S  -

         CAGATTGGTTTCTCCACTCATCGGCTAAAGGATGGGATTCTTTTTGGGATGGTGGGGGCC
    1141 ---------+---------+---------+---------+---------+---------+ 1200
         GTCTAACCAAAGAGGTGAGTAGCCGATTTCCTACCCTAAGAAAAACCCTACCACCCCCGG

a        Q  I  G  F  S  T  H  R  L  K  D  G  I  L  F  G  M  V  G  A  -

         TATGACTGGGGAGGCTCTGTGCTATGGCTTGAAGGAGGCCACCGCCTTTTCCCCCCACGA
    1201 ---------+---------+---------+---------+---------+---------+ 1260
         ATACTGACCCCTCCGAGACACGATACCGAACTTCCTCCGGTGGCGGAAAAGGGGGGTGCT

a        Y  D  W  G  G  S  V  L  W  L  E  G  G  H  R  L  F  P  P  R  -

         ATGGCACTGGAAGACGAGTTCCCCCCTGCACTGCAGAACCATGCAGCCTACCTGGGTTAC
    1261 ---------+---------+---------+---------+---------+---------+ 1320
         TACCGTGACCTTCTGCTCAAGGGGGGACGTGACGTCTTGGTACGTCGGATGGACCCAATG

a        M  A  L  E  D  E  F  P  P  A  L  Q  N  H  A  A  Y  L  G  Y  -

         TCTGTTTCTTCCATGCTTTTGCGGGGTGGACGCCGCCTGTTTCTCTCTGGGGCTCCTCGA
    1321 ---------+---------+---------+---------+---------+---------+ 1380
         AGACAAAGAAGGTACGAAAACGCCCCACCTGCGGCGGACAAAGAGAGACCCCGAGGAGCT

a        S  V  S  S  M  L  L  R  G  G  R  R  L  F  L  S  G  A  P  R  -

         TTTAGACATCGAGGAAAAGTCATCGCCTTCCAGCTTAAGAAAGATGGGGCTGTGAGGGTT
    1381 ---------+---------+---------+---------+---------+---------+ 1440
         AAATCTGTAGCTCCTTTTCAGTAGCGGAAGGTCGAATTCTTTCTACCCCGACACTCCCAA

a        F  R  H  R  G  K  V  I  A  F  Q  L  K  K  D  G  A  V  R  V  -

         GCCCAGAGCCTCCAGGGGGAGCAGATTGGTTCATACTTTGGCAGTGAGCTCTGCCCATTG
    1441 ---------+---------+---------+---------+---------+---------+ 1500
         CGGGTCTCGGAGGTCCCCCTCGTCTAACCAAGTATGAAACCGTCACTCGAGACGGGTAAC

a        A  Q  S  L  Q  G  E  Q  I  G  S  Y  F  G  S  E  L  C  P  L  -

         GATACAGATAGGGATGGAACAACTGATGTCTTACTTGTGGCTGCCCCCATGTTCCTGGGA
    1501 ---------+---------+---------+---------+---------+---------+ 1560
         CTATGTCTATCCCTACCTTGTTGACTACAGAATGAACACCGACGGGGGTACAAGGACCCT

a        D  T  D  R  D  G  T  T  D  V  L  L  V  A  A  P  M  F  L  G  -

         CCCCAGAACAAGGAAACAGGACGTGTTTATGTGTATCTGGTAGGCCAGCAGTCCTTGCTG
    1561 ---------+---------+---------+---------+---------+---------+ 1620
         GGGGTCTTGTTCCTTTGTCCTGCACAAATACACATAGACCATCCGGTCGTCAGGAACGAC

a        P  Q  N  K  E  T  G  R  V  Y  V  Y  L  V  G  Q  Q  S  L  L  -

         ACCCTCCAAGGAACACTTCAGCCAGAACCCCCCCAGGATGCTCGGTTTGGCTTTGCCATG
    1621 ---------+---------+---------+---------+---------+---------+ 1680
         TGGGAGGTTCCTTGTGAAGTCGGTCTTGGGGGGGTCCTACGAGCCAAACCGAAACGGTAC

a        T  L  Q  G  T  L  Q  P  E  P  P  Q  D  A  R  F  G  F  A  M  -

         GGAGCTCTTCCTGATCTGAACCAAGATGGTTTTGCTGATGTGGCTGTGGGGGCGCCTCTG
    1681 ---------+---------+---------+---------+---------+---------+ 1740
         CCTCGAGAAGGACTAGACTTGGTTCTACCAAAACGACTACACCGACACCCCCGCGGAGAC

a        G  A  L  P  D  L  N  Q  D  G  F  A  D  V  A  V  G  A  P  L  -

         GAAGATGGGCACCAGGGAGCACTGTACCTGTACCATGGAACCCAGAGTGGAGTCAGGCCC
    1741 ---------+---------+---------+---------+---------+---------+ 1800
         CTTCTACCCGTGGTCCCTCGTGACATGGACATGGTACCTTGGGTCTCACCTCAGTCCGGG
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a          E  D  G  H  Q  G  A  L  Y  L  Y  H  G  T  Q  S  G  V  R  P  -

           CATCCTGCCCAGAGGATTGCTGCTGCCTCCATGCCACATGCCCTCAGCTACTTTGGCCGA
      1801 ---------+---------+---------+---------+---------+---------+ 1860
           GTAGGACGGGTCTCCTAACGACGACGGAGGTACGGTGTACGGGAGTCGATGAAACCGGCT

a          H  P  A  Q  R  I  A  A  A  S  M  P  H  A  L  S  Y  F  G  R  -

           AGTGTGGATGGTCGGCTAGATCTGGATGGAGATGATCTGGTCGATGTGGCTGTGGGTGCC
      1861 ---------+---------+---------+---------+---------+---------+ 1920
           TCACACCTACCAGCCGATCTAGACCTACCTCTACTAGACCAGCTACACCGACACCCACGG

a          S  V  D  G  R  L  D  L  D  G  D  D  L  V  D  V  A  V  G  A  -

           CAGGGGGCAGCCATCCTGCTCAGCTCCCGGCCCATTGTCCATCTGACCCCATCACTGGAG
      1921 ---------+---------+---------+---------+---------+---------+ 1980
           GTCCCCCGTCGGTAGGACGAGTCGAGGGCCGGGTAACAGGTAGACTGGGGTAGTGACCTC

a          Q  G  A  A  I  L  L  S  S  R  P  I  V  H  L  T  P  S  L  E  -

           GTGACCCCACAGGCCATCAGTGTGGTTCAGAGGGACTGTAGGCGGCGAGGCCAAGAAGCA
      1981 ---------+---------+---------+---------+---------+---------+ 2040
           CACTGGGGTGTCCGGTAGTCACACCAAGTCTCCCTGACATCCGCCGCTCCGGTTCTTCGT

a          V  T  P  Q  A  I  S  V  V  Q  R  D  C  R  R  R  G  Q  E  A  -

           GTCTGTCTGACTGCAGCCCTTTGCTTCCAAGTGACCTCCCGTACTCCTGGTCGCTGGGAT
      2041 ---------+---------+---------+---------+---------+---------+ 2100
           CAGACAGACTGACGTCGGGAAACGAAGGTTCACTGGAGGGCATGAGGACCAGCGACCCTA

a          V  C  L  T  A  A  L  C  F  Q  V  T  S  R  T  P  G  R  W  D  -

           CACCAATTCTACATGAGGTTCACCGCATCACTGGATGAATGGACTGCTGGGGCACGTGCA
      2101 ---------+---------+---------+---------+---------+---------+ 2160
           GTGGTTAAGATGTACTCCAAGTGGCGTAGTGACCTACTTACCTGACGACCCCGTGCACGT

a          H  Q  F  Y  M  R  F  T  A  S  L  D  E  W  T  A  G  A  R  A  -

           GCATTTGATGGCTCTGGCCAGAGGTTGTCCCCTCGGAGGCTCCGGCTCAGTGTGGGGAAT
      2161 ---------+---------+---------+---------+---------+---------+ 2220
           CGTAAACTACCGAGACCGGTCTCCAACAGGGGAGCCTCCGAGGCCGAGTCACACCCCTTA

a          A  F  D  G  S  G  Q  R  L  S  P  R  R  L  R  L  S  V  G  N  -

           GTCACTTGTGAGCAGCTACACTTCCATGTGCTGGATACATCAGATTACCTCCGGCCAGTG
      2221 ---------+---------+---------+---------+---------+---------+ 2280
           CAGTGAACACTCGTCGATGTGAAGGTACACGACCTATGTAGTCTAATGGAGGCCGGTCAC

a          V  T  C  E  Q  L  H  F  H  V  L  D  T  S  D  Y  L  R  P  V  -

           GCCTTGACTGTGACCTTTGCCTTGGACAATACTACAAAGCCAGGGCCTGTGCTGAATGAG
      2281 ---------+---------+---------+---------+---------+---------+ 2340
           CGGAACTGACACTGGAAACGGAACCTGTTATGATGTTTCGGTCCCGGACACGACTTACTC

a          A  L  T  V  T  F  A  L  D  N  T  T  K  P  G  P  V  L  N  E  -

           GGCTCACCCACCTCTATACAAAAGCTGGTCCCCTTCTCAAAGGATTGTGGCCCTGACAAT
      2341 ---------+---------+---------+---------+---------+---------+ 2400
           CCGAGTGGGTGGAGATATGTTTTCGACCAGGGGAAGAGTTTCCTAACACCGGGACTGTTA

a          G  S  P  T  S  I  Q  K  L  V  P  F  S  K  D  C  G  P  D  N  -

           GAATGTGTCACAGACCTGGTGCTTCAAGTGAATATGGACATCAGAGGCTCCAGGAAGGCC
      2401 ---------+---------+---------+---------+---------+---------+ 2460
           CTTACACAGTGTCTGGACCACGAAGTTCACTTATACCTGTAGTCTCCGAGGTCCTTCCGG
```

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a        E  C  V  T  D  L  V  L  Q  V  N  M  D  I  R  G  S  R  K  A   -

     CCATTTGTGGTTCGAGGTGGCCGGCGGAAAGTGCTGGTATCTACAACTCTGGAGAACAGA
2461 ---------+---------+---------+---------+---------+---------+ 2520
     GGTAAACACCAAGCTCCACCGGCCGCCTTTCACGACCATAGATGTTGAGACCTCTTGTCT

a        P  F  V  V  R  G  G  R  R  K  V  L  V  S  T  T  L  E  N  R   -

     AAGGAAAATGCTTACAATACGAGCCTGAGTATCATCTTCTCTAGAAACCTCCACCTGGCC
2521 ---------+---------+---------+---------+---------+---------+ 2580
     TTCCTTTTACGAATGTTATGCTCGGACTCATAGTAGAAGAGATCTTTGGAGGTGGACCGG

a        K  E  N  A  Y  N  T  S  L  S  I  I  F  S  R  N  L  H  L  A   -

     AGTCTCACTCCTCAGAGAGAGAGCCCAATAAAGGTGGAATGTGCCGCCCCTTCTGCTCAT
2581 ---------+---------+---------+---------+---------+---------+ 2640
     TCAGAGTGAGGAGTCTCTCTCTCGGGTTATTTCCACCTTACACGGCGGGGAAGACGAGTA

a        S  L  T  P  Q  R  E  S  P  I  K  V  E  C  A  A  P  S  A  H   -

     GCCCGGCTCTGCAGTGTGGGGCATCCTGTCTTCCAGACTGGAGCCAAGGTGACCTTTCTG
2641 ---------+---------+---------+---------+---------+---------+ 2700
     CGGGCCGAGACGTCACACCCCGTAGGACAGAAGGTCTGACCTCGGTTCCACTGGAAAGAC

a        A  R  L  C  S  V  G  H  P  V  F  Q  T  G  A  K  V  T  F  L   -

     CTAGAGTTTGAGTTTAGCTGCTCCTCTCTCCTGAGCCAGGTCTTTGGGAAGCTGACTGCC
2701 ---------+---------+---------+---------+---------+---------+ 2760
     GATCTCAAACTCAAATCGACGAGGAGAGAGGACTCGGTCCAGAAACCCTTCGACTGACGG

a        L  E  F  E  F  S  C  S  S  L  L  S  Q  V  F  G  K  L  T  A   -

     AGCAGTGACAGCCTGGAGAGAAATGGCACCCTTCAAGAAAACACAGCCCAGACCTCAGCC
2761 ---------+---------+---------+---------+---------+---------+ 2820
     TCGTCACTGTCGGACCTCTCTTTACCGTGGGAAGTTCTTTTGTGTCGGGTCTGGAGTCGG

a        S  S  D  S  L  E  R  N  G  T  L  Q  E  N  T  A  Q  T  S  A   -

     TACATCCAATATGAGCCCCACCTCCTGTTCTCTAGTGAGTCTACCCTGCACCGCTATGAG
2821 ---------+---------+---------+---------+---------+---------+ 2880
     ATGTAGGTTATACTCGGGGTGGAGGACAAGAGATCACTCAGATGGGACGTGGCGATACTC

a        Y  I  Q  Y  E  P  H  L  L  F  S  S  E  S  T  L  H  R  Y  E   -

     GTTCACCCATATGGGACCCTCCCAGTGGGTCCTGGCCCAGAATTCAAAACCACTCTCAGG
2881 ---------+---------+---------+---------+---------+---------+ 2940
     CAAGTGGGTATACCCTGGGAGGGTCACCCAGGACCGGGTCTTAAGTTTTGGTGAGAGTCC

a        V  H  P  Y  G  T  L  P  V  G  P  G  P  E  F  K  T  T  L  R   -

     ACTAACAATGCAAGCTGCATAGTGCAGAACCTGACTGAACCCCCAGGCCCACCTGTGCAT
2941 ---------+---------+---------+---------+---------+---------+ 3000
     TGATTGTTACGTTCGACGTATCACGTCTTGGACTGACTTGGGGGTCCGGGTGGACACGTA

a        T  N  N  A  S  C  I  V  Q  N  L  T  E  P  P  G  P  P  V  H   -

     CCAGAGGAGCTTCAACACACAAACAGACTGAATGGGAGCAATACTCAGTGTCAGGTGGTG
3001 ---------+---------+---------+---------+---------+---------+ 3060
     GGTCTCCTCGAAGTTGTGTGTTTGTCTGACTTACCCTCGTTATGAGTCACAGTCCACCAC

a        P  E  E  L  Q  H  T  N  R  L  N  G  S  N  T  Q  C  Q  V  V   -

     AGGTGCCACCTTGGGCAGCTGGCAAAGGGGACTGAGGTCTCTGTTGGACTATTGAGGCTG
3061 ---------+---------+---------+---------+---------+---------+ 3120
     TCCACGGTGGAACCCGTCGACCGTTTCCCCTGACTCCAGAGACAACCTGATAACTCCGAC
```

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a        R  C  H  L  G  Q  L  A  K  G  T  E  V  S  V  G  L  L  R  L  -

         GTTCACAATGAATTTTTCCGAAGAGCCAAGTTCAAGTCCCTGACGGTGGTCAGCACCTTT
    3121 ---------+---------+---------+---------+---------+---------+ 3180
         CAAGTGTTACTTAAAAAGGCTTCTCGGTTCAAGTTCAGGGACTGCCACCAGTCGTGGAAA

a        V  H  N  E  F  F  R  R  A  K  F  K  S  L  T  V  V  S  T  F  -

         GAGCTGGGAACCGAAGAGGGCAGTGTCCTACAGCTGACTGAAGCCTCCCGTTGGAGTGAG
    3181 ---------+---------+---------+---------+---------+---------+ 3240
         CTCGACCCTTGGCTTCTCCCGTCACAGGATGTCGACTGACTTCGGAGGGCAACCTCACTC

a        E  L  G  T  E  E  G  S  V  L  Q  L  T  E  A  S  R  W  S  E  -

         AGCCTCTTGGAGGTGGTTCAGACCCGGCCTATCCTCATCTCCCTGTGGATCCTCATAGGC
    3241 ---------+---------+---------+---------+---------+---------+ 3300
         TCGGAGAACCTCCACCAAGTCTGGGCCGGATAGGAGTAGAGGGACACCTAGGAGTATCCG

a        S  L  L  E  V  V  Q  T  R  P  I  L  I  S  L  W  I  L  I  G  -

         AGTGTCCTGGGAGGGTTGCTCCTGCTTGCTCTCCTTGTCTTCTGCCTGTGGAAGCTTGGC
    3301 ---------+---------+---------+---------+---------+---------+ 3360
         TCACAGGACCCTCCCAACGAGGACGAACGAGAGGAACAGAAGACGGACACCTTCGAACCG

a        S  V  L  G  G  L  L  L  L  A  L  L  V  F  C  L  W  K  L  G  -

         TTCTTTGCCCATAAGAAAATCCCTGAGGAAGAAAAAAGAGAAGAGAAGTTGGAGCAATGA
    3361 ---------+---------+---------+---------+---------+---------+ 3420
         AAGAAACGGGTATTCTTTTAGGGACTCCTTCTTTTTTCTCTTCTCTTCAACCTCGTTACT

a        F  F  A  H  K  K  I  P  E  E  E  K  R  E  E  K  L  E  Q

         ATGTAGAATAAGGGTCTAGAAAGTCCTCCCTGGCAGCTTTCTTCAAGAGACTTGCATAAA
    3421 ---------+---------+---------+---------+---------+---------+ 3480
         TACATCTTATTCCCAGATCTTTCAGGAGGGACCGTCGAAAGAAGTTCTCTGAACGTATTT

         AGCAGAGGTTTGGGGGCTCAGATGGGACAAGAAGCCGCCTCTGGACTATCTCCCCAGACC
    3481 ---------+---------+---------+---------+---------+---------+ 3540
         TCGTCTCCAAACCCCCGAGTCTACCCTGTTCTTCGGCGGAGACCTGATAGAGGGGTCTGG

         AGCAGCCTGACTTGACTTTTGAGTCCTAGGGATGCTGCTGGCTAGAGATGAGGCTTTACC
    3541 ---------+---------+---------+---------+---------+---------+ 3600
         TCGTCGGACTGAACTGAAAACTCAGGATCCCTACGACGACCGATCTCTACTCCGAAATGG

         TCAGACAAGAAGAGCTGGCACCAAAACTAGCCATGCTCCCACCCTCTGCTTCCCTCCTCC
    3601 ---------+---------+---------+---------+---------+---------+ 3660
         AGTCTGTTCTTCTCGACCGTGGTTTTGATCGGTACGAGGGTGGGAGACGAAGGGAGGAGG

         TCGTGATCCTGGTTCCATAGCCAACACTGGGGCTTTTGTTTGGGGTCCTTTTATCCCCAG
    3661 ---------+---------+---------+---------+---------+---------+ 3720
         AGCACTAGGACCAAGGTATCGGTTGTGACCCCGAAAACAAACCCCAGGAAAATAGGGGTC

         GAATCAATAATTTTTTTGCCTAGGAAAAAAAAAAAGCGGCCGCGAATTCGATATCAAGCT
    3721 ---------+---------+---------+---------+---------+---------  3779
         CTTAGTTATTAAAAAAACGGATCCTTTTTTTTTTTCGCCGGCGCTTAAGCTATAGTTCGA
```

(2) INFORMATION FOR SEQ ID NO. 3:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 143 base pairs
(B) TYPE: nucleic acid and amino acid
(C) STRANDEDNESS: double
(D) TOPOLOGY: linear

(iii) MOLECULAR TYPE: cDNA
(vi) ORIGINAL SOURCE:
(A) ORGANISM: human
(B) CELLTYPE: chondrocyte

(xi) SEQUENCE DESCRIPTION: SEQ ID NO. 3:

```
         NdeI
           |
     GGGGCATATGGTTCAGAACCTGGGTTGCTACGTTGTTTCCGGTCTGATCATCTCCGCTCT
   1 ---------+---------+---------+---------+---------+---------+ 60
     CCCCGTATACCAAGTCTTGGACCCAACGATGCAACAAAGGCCAGACTAGTAGAGGCGAGA

b     G  H  M  V  Q  N  L  G  C  Y  V  V  S  G  L  I  I  S  A  L -

     GCTGCCGGCTGTTGCTCACGGTGGTAACTACTTCCTAAGCTTGTCCCAGGTTATCAGCGG
  61 ---------+---------+---------+---------+---------+---------+ 120
     CGACGGCCGACAACGAGTGCCACCATTGATGAAGGATTCGAACAGGGTCCAATAGTCGCC

b     L  P  A  V  A  H  G  G  N  Y  F  L  S  L  S  Q  V  I  S  G -

                BamHI
                  |
     CCTGGTGCCGCGCGGATCCCCCC
 121 ---------+---------+--- 143
     GGACCACGGCGCGCCTAGGGGGG

b     L  V  P  R  G  S  P   -
```

CLAIMS

1. A recombinant or isolated integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or homologues or fragments thereof having similar biological activity.

2. A process of producing a recombinant integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or homologues or fragments thereof having similar biological activity, which process comprises the steps of

a) isolating a polynucleotide comprising a nucleotide sequence coding for an integrin subunit α10, or homologues or fragments thereof having similar biological activity,

b) constructing an expression vector comprising the isolated polynucleotide,

c) transforming a host cell with said expression vector,

d) culturing said transformed host cell in a culture medium under conditions suitable for expression of integrin subunit α10, or homologues or fragments thereof having similar biological activity, in said transformed host cell, and, optionally,

e) isolating the integrin subunit α10, or homologues or fragments thereof having similar biological activity, from said transformed host cell or said culture medium.

3. A process of providing an integrin subunit α10, or homologues or fragments thereof having similar biological activity, whereby said subunit is isolated from a cell in which it is naturally present.

4. An isolated polynucleotide comprising a nucleotide coding for an integrin subunit α10, or for homologues or fragments thereof, which polynucleotide comprises the nucleotide sequence shown in SEQ ID No. 1 or SEQ ID No. 2 or suitable parts thereof.

5. An isolated polynucleotide or oligonucleotide which hybridises to a DNA or RNA encoding an integrin subunit α10 or homologues or fragments thereof, wherein said polynucleotide or oligonucleotide fails to hybridise to a DNA or RNA encoding an integrin subunit α1.

6. A vector comprising a polynucleotide or oligonucleotide coding for an integrin subunit α10, or homologues or fragments thereof, which polynucleotide or oligonucleotide comprises the nucleotide sequence shown in SEQ ID No. 1 or SEQ ID No. 2 or parts thereof.

7. A vector comprising a polynucleotide or oligonucleotide which hybridises to a DNA or RNA encoding an integrin subunit α10 or homologues or fragments thereof, wherein said polynucleotide or oligonucleotide fails to hybridise to a DNA or RNA encoding an integrin subunit α1.

8. A cell containing the vector as defined in any one of claims 6 and 7.

9. A cell generated by the process in claim 2, in which a polynucleotide or oligonucleotide coding for an integrin subunit α10, or homologues or fragments thereof, which polynucleotide or oligonucleotide comprises the nucleotide sequence shown in SEQ ID No. 1 or SEQ ID No. 2 or parts thereof has been stably integrated in the cell genome.

10. Binding entities having the capability of binding specifically to integrin subunit α10 comprising the amino acid sequence of SEQ ID No. 1 or SEQ ID No. 2, or to homologues or fragments thereof.

11. Binding entities according to claim 10, which are chosen from the group comprising proteins, peptides, carbohydrates, lipids, natural integrin binding ligands, polyclonal and monoclonal antibodies, and fragments thereof.

12. A recombinant or isolated integrin heterodimer comprising a subunit α10 and a subunit β, in which the

subunit $\alpha10$ comprises the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, and homologues and fragments thereof having similar biological activity.

13. A recombinant or isolated integrin heterodimer according to claim 12, wherein the subunit β is $\beta1$.

14. A process of producing a recombinant integrin heterodimer comprising a subunit $\alpha10$ and a subunit β, in which the subunit $\alpha10$ comprises the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, and homologues and fragments thereof, which process comprises the steps of

a) isolating one polynucleotide comprising a nucleotide sequence coding for a subunit $\alpha10$ of an integrin heterodimer and, optionally, another polynucleotide comprising a nucleotide sequence coding for a subunit β of an integrin heterodimer, or polynucleotides or oligonucleotides coding for homologues or fragments thereof having similar biological activity,

b) constructing an expression vector comprising said isolated polynucleotide coding for said subunit $\alpha10$ optionally in combination with an expression vector comprising said isolated nucleotide coding for said subunit β,

c) transforming a host cell with said expression vector or vectors,

d) culturing said transformed host cell in a culture medium under conditions suitable for expression of an integrin heterodimer comprising a subunit $\alpha10$ and a subunit β, or homologues or fragments thereof having similar biological activity, in said transformed host cell, and, optionally,

e) isolating the integrin heterodimer comprising a subunit $\alpha10$ and a subunit β, or homologues or fragments thereof having similar biological activity, or the $\alpha10$ subunit thereof from said transformed host cell or said culture medium.

15. A process of providing an integrin heterodimer comprising a subunit $\alpha10$ and a subunit β, or homologues

or fragments thereof having similar biological activity, whereby said integrin heterodimer is isolated from a cell in which it is naturally present.

16. A cell containing a first vector, said first vector comprising a polynucleotide or oligonucleotide coding for a subunit α10 of an integrin heterodimer, or for homologues or parts thereof having similar biological activity, which polynucleotide or oligonucleotide comprises the nucleotide sequence shown in SEQ ID No. 1 or SEQ ID No. 2 or parts thereof, and a second vector, said second vector comprising a polynucleotide or oligonucleotide coding for a subunit β of an integrin heterodimer, or for homologues or fragments thereof.

17. Binding entities having the capability of binding specifically to the integrin heterodimer comprising a subunit α10 and a subunit β, or to homologues or fragments thereof, or a subunit α10 thereof, having similar biological activity.

18. Binding entities according to claim 17, wherein the subunit β is β1.

19. Binding entities according to claim 17 or 18, which are chosen among the group comprising proteins, peptides, carbohydrates, lipids, natural integrin binding ligands, and fragments thereof.

20. A fragment of the integrin subunit α10, which fragment is a peptide chosen from the group comprising peptides of the cytoplasmic domain, the I-domain and the spliced domain.

21. A fragment according to claim 20, which is a peptide comprising the amino acid sequence KLGFFAHKKIPEEEKREEKLEQ.

22. A fragment according to claim 20, which comprises the amino acid sequence from about amino acid No. 952 to about amino acid no. 986 of SEQ ID No. 1.

23. A fragment according to claim 20, which is a peptide comprising the amino acid sequence from about

amino acid No. 140 to about amino acid no. 337 of SEQ ID No. 1.

24. A method of producing a fragment of the integrin subunit α10 as defined in any one of claims 20-23, which method comprises a sequential addition of amino acids containing protective groups.

25. A polynucleotide or oligonucleotide coding for a fragment of the integrin subunit α10 as defined in any one of claims 20-23.

26. Binding entities having the capability of binding specifically to a fragment of the human integrin subunit α10 as defined in any one of claims 20-23.

27. Binding entities according to claim 26, which are chosen from the group comprising proteins, peptides, carbohydrates, lipids, natural integrin binding ligands, and fragments thereof.

28. A process of using an integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or an integrin heterodimer comprising said subunit α10 and a subunit β, or a homologue or fragment of said integrin or subunit having similar biological activity, as a marker or target molecule of cells or tissues expressing said integrin subunit α10, which cells or tissues are of animal including human origin.

29. A process according to claim 28, whereby said fragment is a peptide chosen from the group comprising peptides of the cytoplasmic domain, the I-domain and the spliced domain.

30. A process according to claim 29, whereby said fragment is a peptide comprising the amino acid sequence KLGFFAHKKIPEEEKREEKLEQ.

31. A process according to claim 29, whereby said fragment comprises the amino acid sequence from about amino acid no. 952 to about amino acid no. 986 of SEQ ID No. 1.

32. A process according to claim 29, whereby said fragment comprises the amino acid sequence from about

amino acid no. 140 to about amino acid no. 337 of SEQ ID No. 1.

33. A process according to claim 28, whereby the subunit β is β1.

34. A process according to claim 28, whereby said cells are chosen from the group comprising chondrocytes, smooth muscle cells, endothelial cells, osteoblasts and fibroblasts.

35. A process according to any one of claims 28-34, which process is used during pathological conditions involving said subunit α10.

36. A process according to claim 35, which pathological conditions comprise damage of cartilage.

37. A process according to claim 36, which pathological conditions comprise trauma, rheumatoid arthritis and osteoarthritis.

38. A process according to any one of claims 28-34, which is a process for detecting the formation of cartilage during embryonal development.

39. A process according to any one of claims 28-34, which is a process for detecting physiological or therapeutic reparation of cartilage.

40. A process according to any one of claims 28-34, which is a process for selection and analysis, or for sorting, isolating or purification of chondrocytes.

41. A process according to any one of claims 28-34, which is a process for detecting regeneration of cartilage or chondrocytes during transplantation of cartilage or chondrocytes.

42. A process according to any one of claims 28-34, which is a process for in vitro studies of differentiation of chondrocytes.

43. A process of using binding entities having the capability of binding specifically to an integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or an integrin heterodimer comprising said subunit α10 and a subunit β, or to homo-

logues or fragments thereof having similar biological activity, as markers or target molecules of cells or tissues expressing said integrin subunit α10, which cells or tissues are of animal including human origin.

44. A process according to claim 43, whereby said fragment is a peptide chosen from the group comprising peptides of the cytoplasmic domain, the I-domain and the spliced domain.

45. A process according to claim 43, whereby said fragment is a peptide comprising the amino acid sequence KLGFFAHKKIPEEEKREEKLEQ.

46. A process according to claim 43, whereby said fragment comprises the amino acid sequence from about amino acid no. 952 to about amino acid no. 986 of SEQ ID No. 1.

47. A process according to claim 43, whereby said fragment comprises the amino acid sequence from about amino acid no. 140 to about amino acid No. 337 of SEQ ID No. 1.

48. A process according to claim 43, whereby the subunit β is β1.

49. A process according to any one of claims 43-48, which is a process for detecting the presence of an integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or of an integrin heterodimer comprising said subunit α10 and a subunit β, or of homologues or fragments thereof having similar biological activity.

50. A process according to any one of claims 43-48, which process is a process for determining the differentiation-state of cells during embryonic development, angiogenesis, or development of cancer.

51. A process for detecting the presence of an integrin subunit α10, or of a homologue or fragment of said integrin subunit having similar biological activity, on cells, whereby a polynucleotide or oligonucleotide chosen from the group comprising a polynucleotide or oligo-

nucleotide shown in SEQ ID No. 1 is used as a marker under hybridisation conditions wherein said polynucleotide or oligonucleotide fails to hybridise to a DNA or RNA encoding an integrin subunit α1.

52. A process according to claim 51, whereby said cells are chosen from the group comprising chondrocytes, smooth muscle cells, endothelial cells, osteoblasts and fibroblasts.

53. A process according to claim 51, whereby said fragment is a peptide chosen from the group comprising peptides of the cytoplasmic domain, the I-domain and the spliced domain.

54. A process according to claim 53, whereby said fragment is a peptide comprising the amino acid sequence KLGFFAHKKIPEEEKREEKLEQ.

55. A process according to claim 53, whereby said fragment comprises the amino acid sequence from about amino acid No. 952 to about amino acid no. 986 of SEQ ID No. 1.

56. A process according to claim 53, whereby said fragment comprises the amino acid sequence from about amino acid No. 140 to about amino acid No. 337 of SEQ ID No. 1.

57. A process according to any one of claims 43-48, which is a process for determining the differentiation-state of cells during development, in pathological conditions, in tissue regeneration or in therapeutic and physiological reparation of cartilage.

58. A process according to claim 57, wherein the pathological conditions are any pathological conditions involving the integrin subunit α10.

59. A process according to claim 58, whereby said pathological conditions are rheumatoid arthritis, osteoarthrosis or cancer.

60. A process according to claim 57, whereby said cells are chosen from the group comprising chondrocytes,

smooth muscle cells, endothelial cells, osteoblasts and fibroblasts.

61. A process for determining the differentiation-state of cells during development, in pathological conditions, in tissue regeneration and in therapeutic and physiological reparation of cartilage, whereby a polynucleotide or oligonucleotide chosen from the nucleotide sequence shown in SEQ ID No. 1 is used as a marker under hybridisation conditions wherein said polynucleotide or oligonucleotide fails to hybridise to a DNA or RNA encoding an integrin subunit α1.

62. A process according to claim 61, whereby said polynucleotide or oligonucleotide is a polynucleotide or oligonucleotide coding for a peptide chosen from the group comprising peptides of the cytoplasmic domain, the I-domain and the spliced domain.

63. A process according to claim 62, whereby said polynucleotide or oligonucleotide is a polynucleotide or oligonucleotide coding for a peptide comprising the amino acid sequence KLGFFAHKKIPEEEKREEKLEQ.

64. A process according to claim 62, whereby said peptide comprises the amino acid sequence from about amino acid no. 952 to about amino acid no. 986 of SEQ ID No. 1.

65. A process according to claim 62, whereby said peptide comprises the amino acid sequence from about amino acid no. 140 to about amino acid no. 337 of SEQ ID No. 1.

66. A process according to claim 61, whereby said pathological conditions are any pathological conditions involving the integrin subunit α10.

67. A process according to claim 66, whereby said pathological conditions are rheumatoid arthritis, osteoarthrosis or cancer.

68. A process according to claim 66, whereby said pathological conditions are atherosclerosis or inflammation.

69. A process according to any one of claims 61-68, whereby said cells are chosen from the group comprising chondrocytes, smooth muscle cells, endothelial cells, osteoblasts and fibroblasts.

70. A pharmaceutical composition comprising as an active ingredient a pharmaceutical agent or an antibody which is capable of using an integrin heterodimer comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit α10 having similar biological activity, as a target molecule.

71. A pharmaceutical composition according to claim 70, for use in stimulating, inhibiting or blocking the formation of cartilage, bone or blood vessels.

72. A pharmaceutical composition according to claim 70, for use in preventing adhesion between tendon/ligaments and the surrounding tissue after infection, inflammation and after surgical intervention where adhesion impairs the function of the tissue.

73. A vaccine comprising as an active ingredient an integrin heterodimer comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit α10, or DNA or RNA coding for said integrin subunit α10.

74. Use of the integrin subunit α10 as a marker or target in transplantation of cartilage or chondrocytes.

75. A method of using binding entities having the capability of binding specifically to an integrin subunit α10 comprising the amino acid sequence shown in SEQ ID No. 1 or SEQ ID No. 2, or an integrin heterodimer comprising said subunit α10 and a subunit β, or to homologues or fragments thereof having similar biological activity, for promoting adhesion of chondrocytes and/or osteoblasts to surfaces of implants to stimulate osseointegration.

76. Use of an integrin heterodimer comprising an integrin subunit α10 and a subunit β, or the subunit α10

thereof, or a homologue or fragment of said integrin or subunit α10 having similar biological activity, as a target for anti-adhesive drugs or molecules in tendon, ligament, skeletal muscle or other tissues where adhesion impairs the function of the tissue.

77. A method of stimulating, inhibiting or blocking the formation of cartilage or bone, comprising administration to a subject a suitable amount of a pharmaceutical agent or an antibody which is capable of using an integrin heterodimer comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit α10 having similar biological activity, as a target molecule.

78. A method of preventing adhesion between tendon/ ligaments and the surrounding tissue after infection, inflammation and after surgical intervention where adhesion impairs the function of the tissue, comprising administration to a subject a suitable amount of a pharmaceutical agent or an antibody which is capable of using a integrin heterodimer comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit α10 having similar biological activity, as a target molecule.

79. A method of stimulating extracellular matrix synthesis and repair by activation or blockage of an integrin heterodimer comprising a subunit α10 and a subunit β, or of the subunit α10 thereof, or of a homologue or fragment of said integrin or subunit α10 having similar biological activity.

80. A method of in vitro detecting the presence of integrin binding entities, comprising interaction of an integrin heterodimer comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit, with a sample, thereby causing said integrin, subunit α10, or homologue or fragment thereof having similar biological activity,

to modulate the binding to its natural ligand or other integrin binding proteins present in said sample.

81. A method of in vitro studying consequences of the interaction of a human heterodimer integrin comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit, with an integrin binding entity and thereby initiate a cellular reaction.

82. A method according to claim 81, whereby the consequences of said interactions are measured as alterations in cellular functions.

83. A method of using DNA or RNA encoding an integrin subunit α10 or homologues or fragments thereof as a target molecule.

84. A method according to claim 83, whereby a polynucleotide or oligonucleotide hybridises to the DNA or RNA encoding an integrin subunit α10 or homologues or fragments thereof and whereby said polynucleotide or oligonucleotide fails to hybridise to a DNA or RNA encoding en integrin subunit α1.

85. A method of using a human heterodimer integrin comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit, or a DNA or RNA encoding an integrin subunit α10 or homologues or fragments thereof, as a marker or target molecule during angiogenesis.

86. A pharmaceutical composition comprising as an active ingredient a pharmaceutical agent or an antibody which is capable of stimulating cell surface expression of an integrin heterodimer comprising a subunit α10 and a subunit β, or the subunit α10 thereof, or a homologue or fragment of said integrin or subunit α10 having similar biological activity.

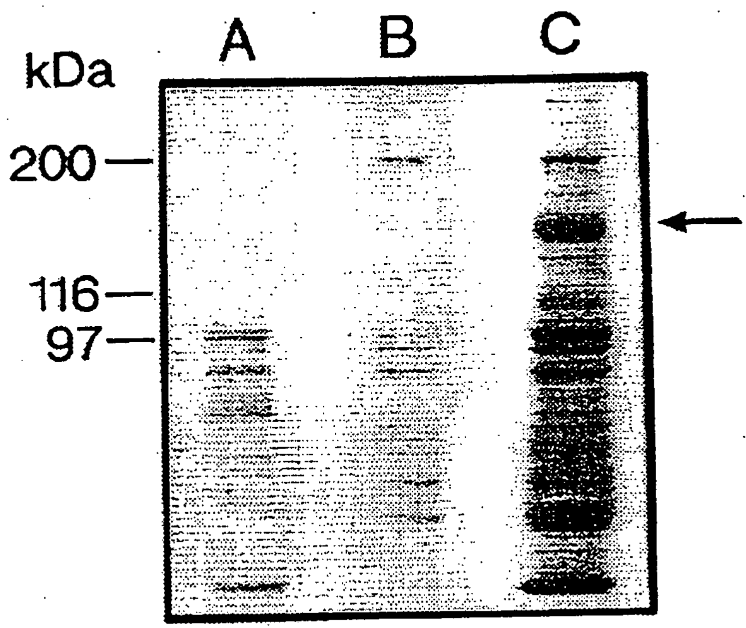


FIGURE 1

Peptide	Amino acid sequence
1	DNTAQTSAYIQYEPHHSI
2	GPGHWDR
3	AAFDGSGQR
4	FAMGALPD
5	FTASLDEWTTAAR
6	VDASFRPQGXLAP

FIGURE 2

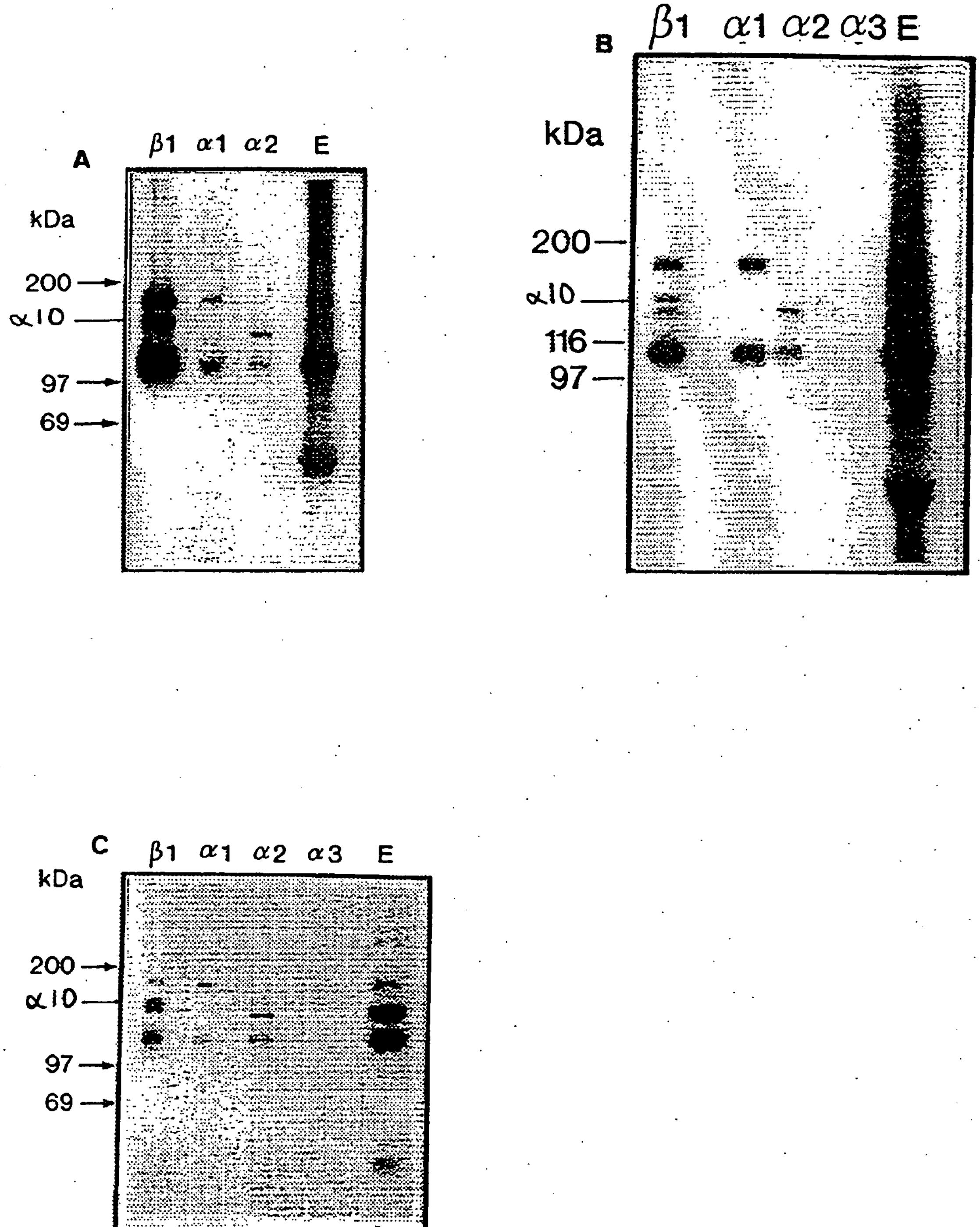

A
β1 α1 α2 E
kDa
200
α10
97
69
B
β1 α1 α2 α3 E
kDa
200
α10
116
97
C
β1 α1 α2 α3 E
kDa
200
α10
97
69

FIGURE 3

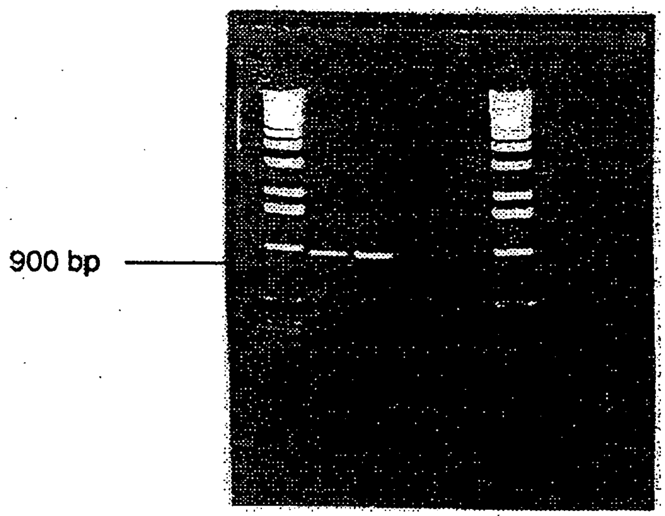


FIGURE 4

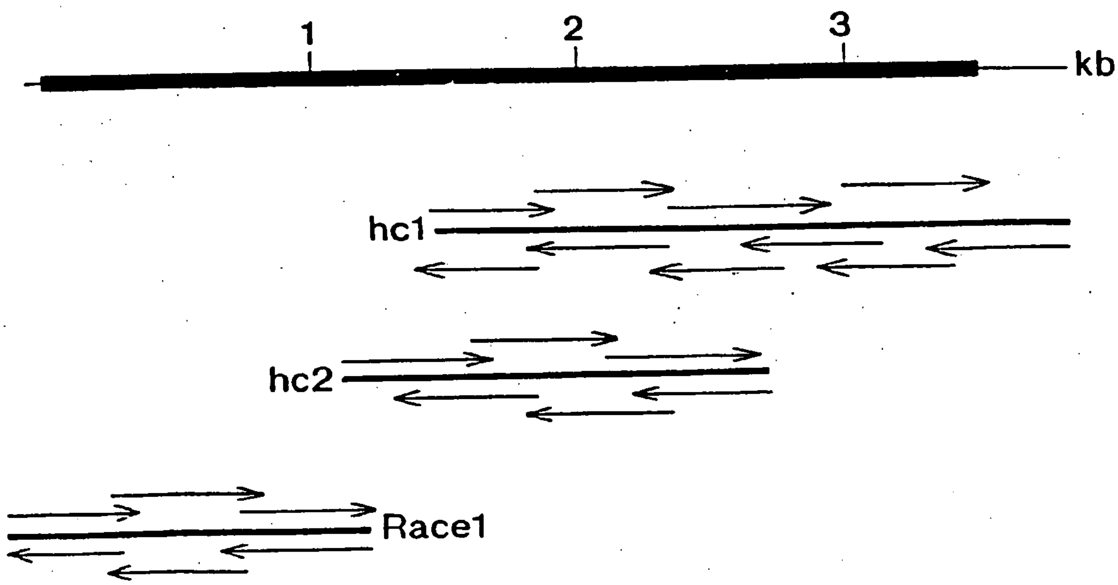


FIGURE 5

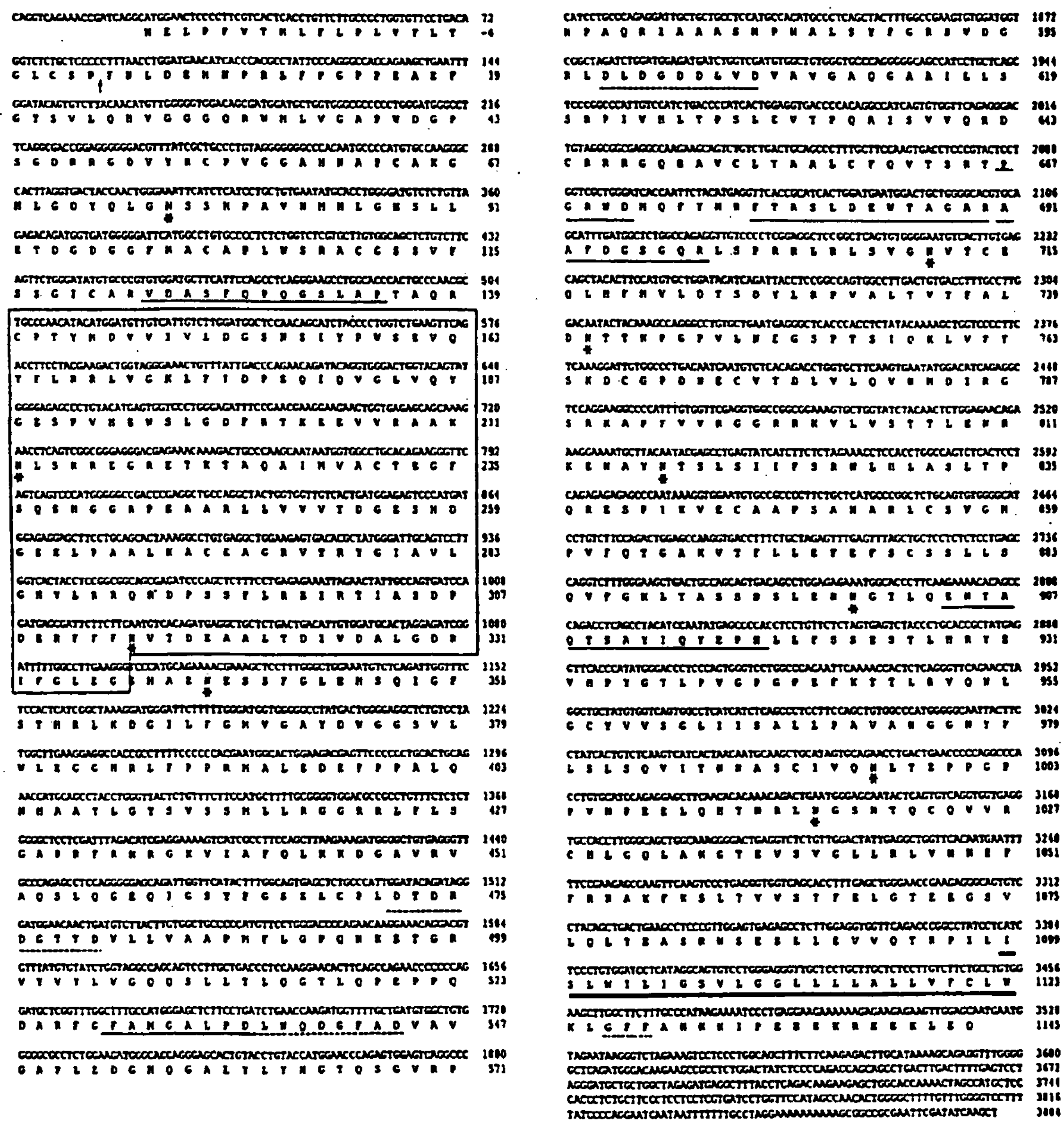


FIGURE 6

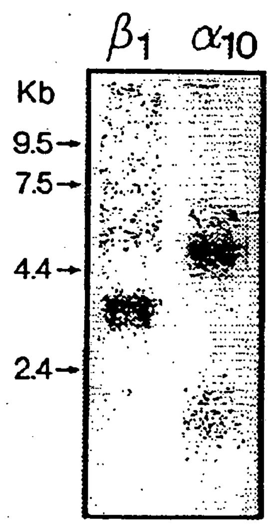
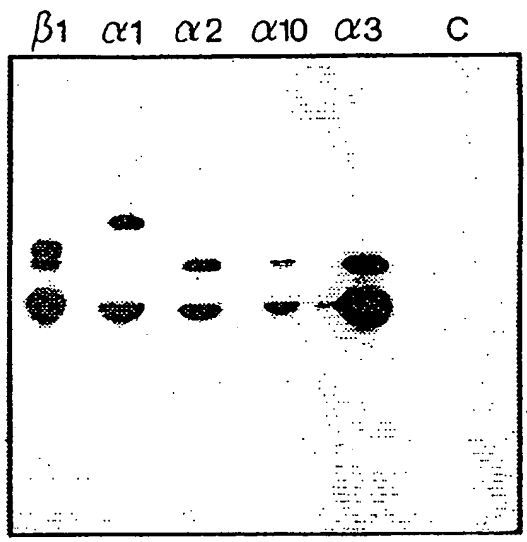


FIGURE 7

A



B

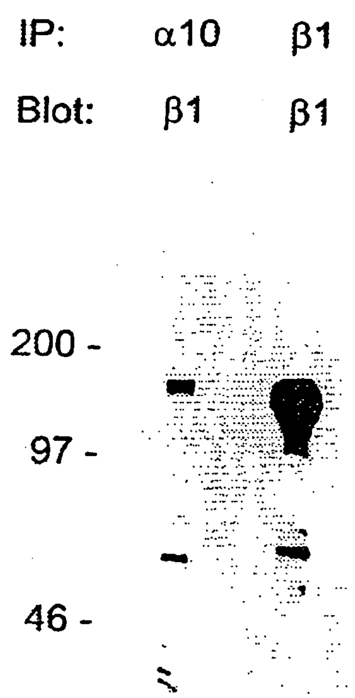


FIGURE 8

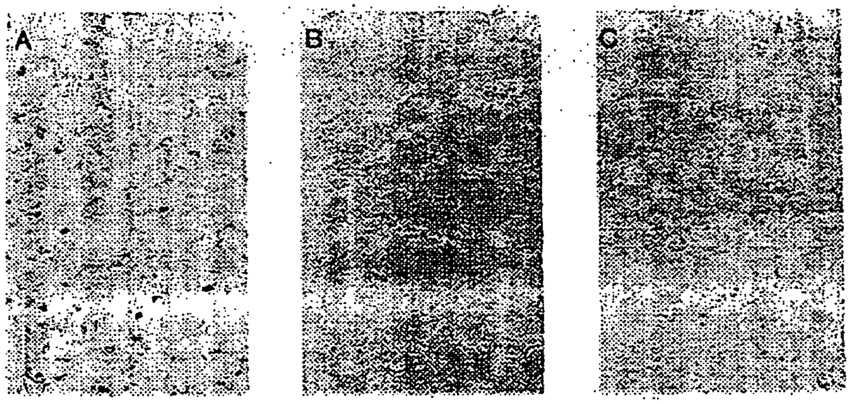


FIGURE 9

A

B

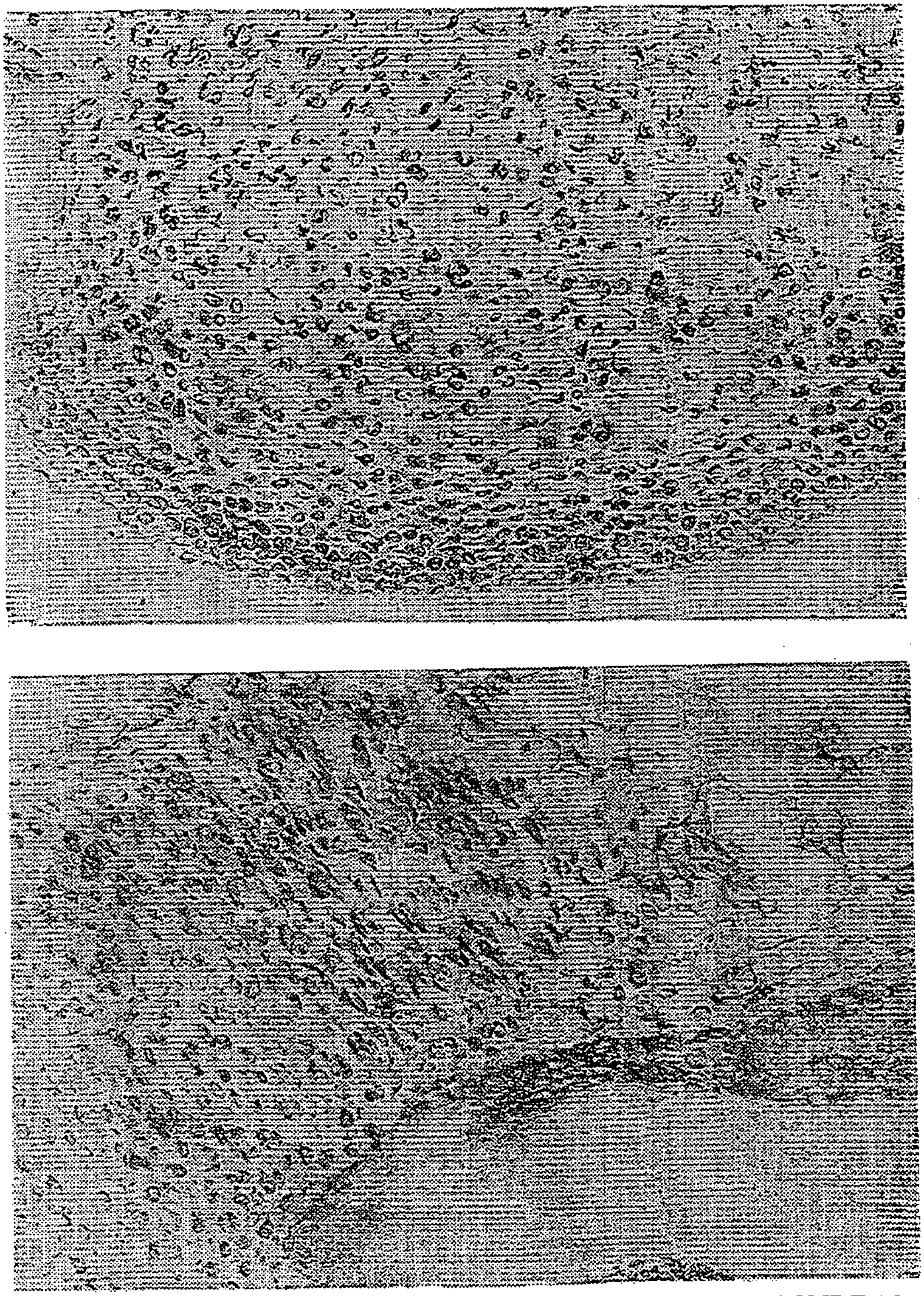
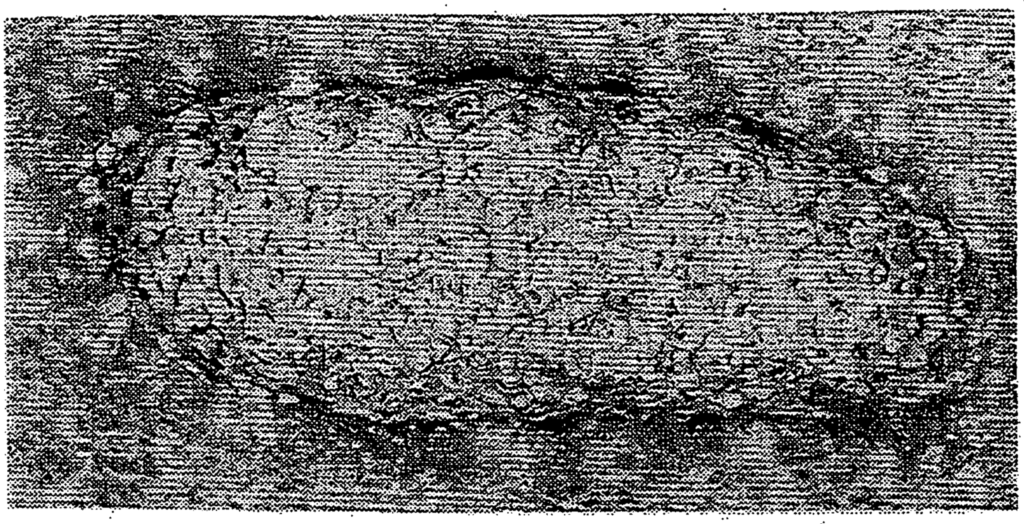
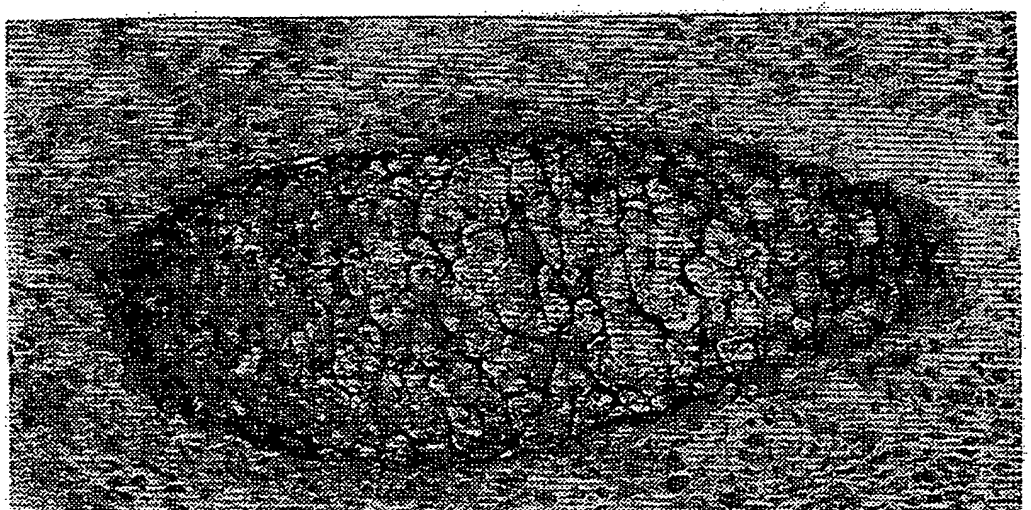


FIGURE 10

A



B



C

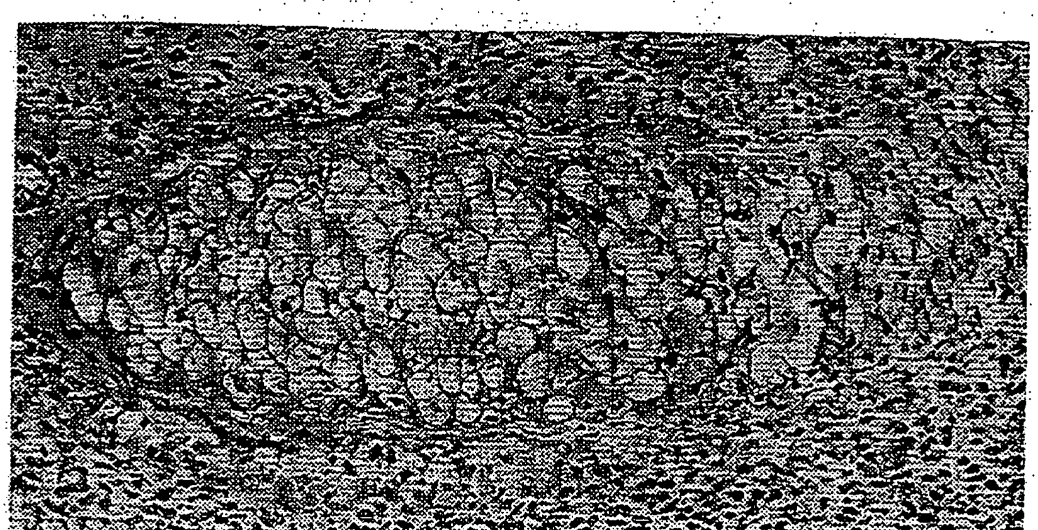


FIGURE 11

Human RNA Master blot

Tissue	α10 expression	Tissue	α10 expression
Aorta	++++	Thyroid gland	-
Trachea	+	Salivary gland	-
Lung	++	Spleen	-
Fetal lung	++	Fetal spleen	-
Kidney	++	Thymus	-
Fetal kidney	(+)	Fetal thymus	-
Heart	(+)	Peripherial leucocyte	-
Fetal heart	++	Lymph node	-
Spinal cord	++	Appendix	-
Mammary gland	(+)	Placenta	-
Bone marrow	(+)	Whole brain	-
Small intestine	(+)	Fetal brain	-
Skeletal muscle	-	Amygdala	-
Liver	-	Caudate nucleus	-
Fetal liver	-	Cerebellum	-
Colon	-	Cerebral cortex	-
Bladder	-	Frontal lobe	-
Uterus	-	Hippocampus	-
Prostate	-	Medulla oblongata	-
Stomach	-	Occipitial lobe	-
Testis	-	Putamen	-
Ovary	-	Substantia nigra	-
Pancreas	-	Temporal lobe	-
Piutiatary gland	-	Thalamus	-
Adrenal gland	-	Subthalamic nucleus	-

FIGURE 12

A

B

C

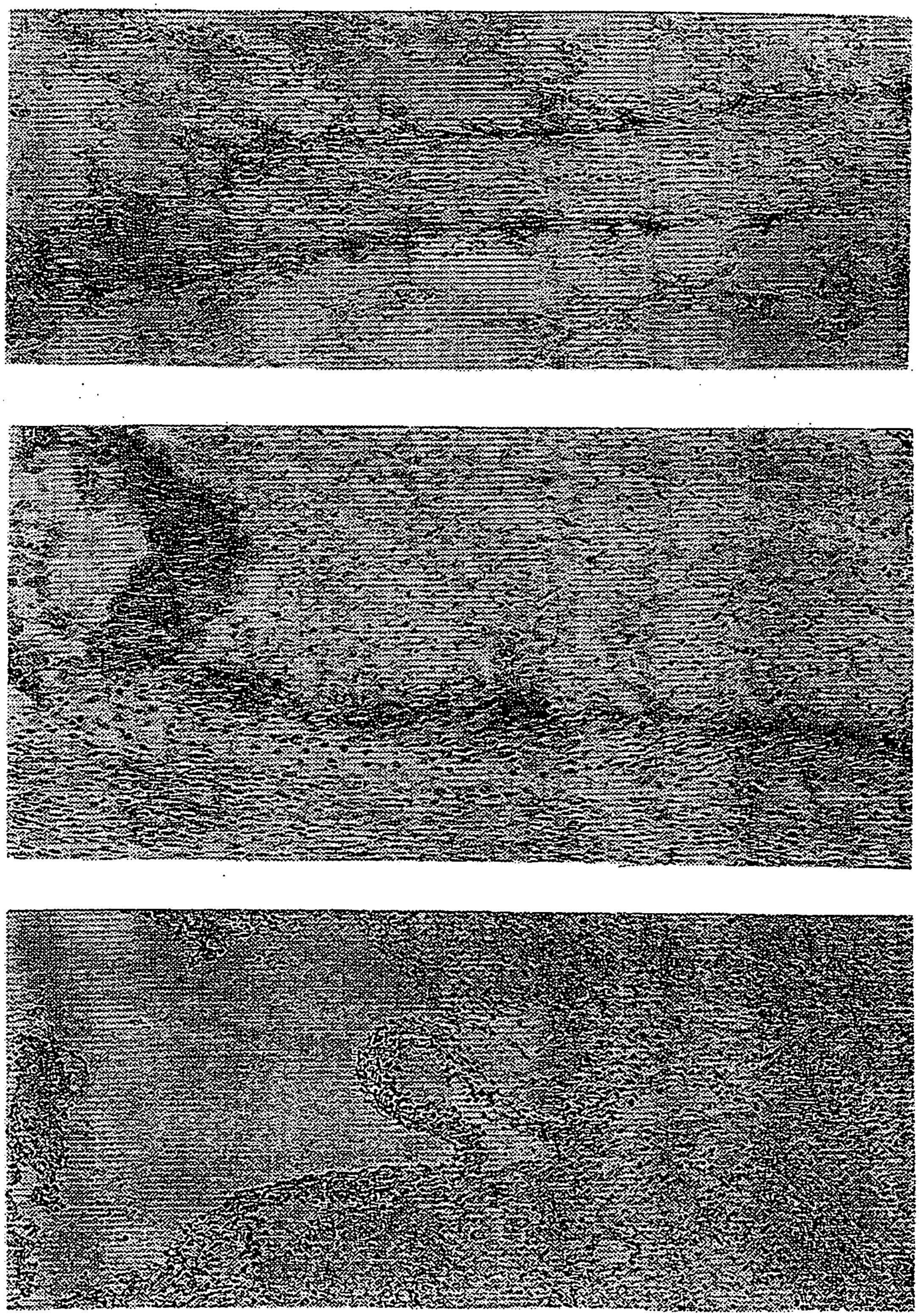


FIGURE 13

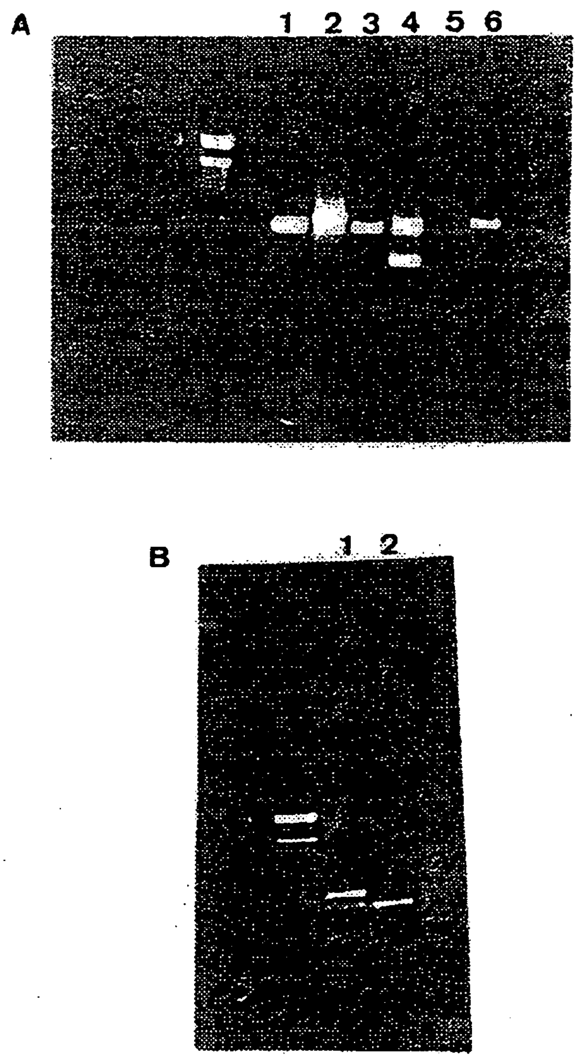


FIGURE 14

```
TGNTHHMKCMCACGAKMGWSAKGNCCGAKGGTKGKGVAAVGTGACARAGCTNGMNAAAARANGAAGTATGACCWGTGGGC 80
 ?  ?  ?  ?  R  ?  ?  ?  P  ?  V  ?  ?  ?  D  ?  A  ?  ?  K  ?  K  Y  D  ?  W  A
  ?  ?  ?  H  ?  ?  ?  ?  R  ?  ?  ?  ?  V  T  ?  L  ?  K  ?  ?  S  H  T  ?  G
V  ?  ?  ?  T  ?  ?  ?  ?  ?  G  ?  ?  ?  .  Q  S  ?  ?  K  ?  E  V  .  P  V  G

CRAGATAGMKAMDAAGCNGMSAGKTRAMGGACGATGGNCCMGCCAAVCGABWGGNAAHTBCGGCNWDCARNGTCCAAATK 160
 ?  I  ?  ?  K  ?  ?  ?  ?  G  R  W  ?  ?  Q  ?  ?  G  ?  ?  G  ?  Q  ?  P  N
P  R  .  ?  ?  S  ?  ?  ?  ?  D  D  G  P  A  ?  R  ?  ?  ?  ?  A  ?  ?  V  Q  ?
 ?  D  ?  ?  ?  A  ?  ?  .  R  T  M  ?  ?  P  ?  ?  ?  ?  ?  R  ?  ?  ?  S  K  ?

SANKTCSCAGGAACCMACGGANTGGCTCGCARCCCDTAGGGATCAGGKACGATGRCTCSCCGRNSKACTCSGNKTGATWA 240
?  ?  ?  R  N  ?  R  ?  G  S  Q  P  ?  G  I  R  ?  D  ?  S  P  ?  ?  S  T  .  ?
 ?  ?  ?  G  T  ?  G  ?  A  R  ?  P  .  G  S  G  T  M  ?  ?  R  ?  T  ?  ?  D  ?
 ?  S  Q  E  P  T  ?  W  L  A  ?  ?  R  D  Q  ?  R  ?  L  ?  ?  ?  L  ?  ?  I

ATCGMNWGTMGGMAGGCGGMGGAATTRWAAAGTANTGGTMGAMAKATGWGVMGGAMATGATRRGTMGACTVTHVMGGVAK 320
 I  ?  ?  R  ?  A  ?  E  L  ?  S  ?  G  R  ?  M  ?  R  ?  .  ?  V  D  ?  ?  G  ?
  S  ?  V  G  R  R  R  N  ?  K  V  ?  V  ?  ?  ?  ?  G  ?  D  ?  ?  ?  ?  ?  ?
N  R  ?  ?  ?  G  G  G  I  ?  K  ?  W  ?  ?  ?  ?  ?  ?  M  ?  ?  R  L  ?  R  ?

VTAKSGGTACAGGCGAAKACARGRAKGTGTCTGAGGAADTCAGNAGGACAAMMTTGCCGAAGTCMGGACTTAGKATRGAT 400
 ?  ?  Y  R  R  ?  Q  ?  ?  V  .  G  ?  Q  ?  D  ?  ?  A  E  V  R  T  .  ?  ?
?  ?  C  T  G  E  ?  ?  ?  V  S  E  E  ?  ?  R  T  ?  L  P  K  S  G  L  ?  ?  D
 ?  ?  V  Q  A  ?  T  ?  ?  C  L  R  ?  S  ?  G  Q  ?  C  R  S  ?  D  L  ?  ?  I

ACGAANCKTRGATCTTAMADGGGGGNKAGCGAGTGCSTAAACGVARATRGGNSWGTCTACTTMAACNCCAAGNGDGGACA 480
Y  E  ?  ?  I  L  ?  G  G  ?  R  V  ?  K  R  ?  ?  ?  ?  L  L  ?  ?  Q  ?  ?  T
 T  ?  ?  ?  S  ?  ?  G  ?  S  E  C  ?  N  ?  ?  ?  ?  V  Y  ?  N  ?  K  ?  G  H
  R  ?  ?  D  L  ?  ?  G  ?  A  S  A  .  T  ?  ?  G  ?  S  T  ?  T  P  ?  ?  D

TTTACTAGASGAGGAGAGTAGCCAGATCACDTGAGATGATCTAAKGTGGGGTCCCGTTGCCAGTATATGAGAGGACTGGT 560
 F  T  R  ?  G  E  .  P  D  H  ?  R  .  S  ?  V  G  S  R  C  Q  Y  M  R  G  L  V
  L  L  ?  E  E  S  S  Q  I  T  .  D  D  L  ?  W  G  P  V  A  S  I  .  E  D  W
I  Y  .  ?  R  R  V  A  R  S  ?  E  M  I  .  ?  G  V  P  L  P  V  Y  E  R  T  G

TCGGCAGACATWGATGCTCTTTGCTGACTCACATATTGTTGCCVTGAGKATGATCAGATACGATCTGWTGTCCCTCATCA 640
 R  Q  T  ?  M  L  F  A  D  S  H  I  V  A  ?  ?  M  I  R  Y  D  L  ?  S  L  I
F  G  R  H  ?  C  S  L  L  T  H  I  L  L  P  .  ?  .  S  D  T  I  ?  C  P  S  S
 S  A  D  I  D  A  L  C  .  L  T  Y  C  C  ?  E  ?  D  Q  I  R  S  ?  V  P  H  H

TGAATSTGRGCCGTGATGCTAATGAGATTCGCCTATGATGGAACAAGAGACTTHTGCTACAGCAGGCGAATGAAGGTTTC 720
M  N  ?  ?  R  D  A  N  E  I  R  L  .  W  N  K  R  L  ?  L  Q  Q  A  N  E  G  F
 .  ?  ?  A  V  M  L  M  R  F  A  Y  D  G  T  R  D  ?  C  Y  S  R  R  M  K  V  S
  E  ?  ?  P  .  C  .  .  D  S  P  M  M  E  Q  E  T  ?  A  T  A  G  E  .  R  F

TAGAGTAGGAGTCTCAGGAGGAGAGAAACTGTGGACCTGGAGGACCAGGGACTCCAGGAGGAAGTWGCCACAACTGGCTT 800
 .  S  R  S  L  R  R  R  E  T  V  D  L  E  D  Q  G  L  Q  E  E  V  A  T  T  G  L
  R  V  G  V  S  G  G  E  K  L  W  T  W  R  T  R  D  S  R  R  K  ?  P  Q  L  A
L  E  .  E  S  Q  E  E  R  N  C  G  P  G  G  P  G  T  P  G  G  S  ?  H  N  W  L

GMAGTTTCGGCTCCGATCCTGATACWGGCTCGTCCTTVGAGTTATCCCCCTCTCTTGCTGGATGGCTCAGAAATGCCTGG 880
 ?  F  R  L  R  S  .  Y  ?  L  V  L  ?  V  I  P  L  S  C  W  M  A  Q  K  C  L
?  S  P  G  S  D  P  D  T  G  S  S  ?  E  L  S  P  S  L  A  G  W  L  R  N  A  W
 ?  V  S  A  P  I  L  I  ?  A  R  P  ?  S  Y  P  P  L  L  L  D  G  S  E  M  P  G

ACCTTTTCATCCCCACTGGACAAACTAGGCGTCTGGCGTTGTGGCCCTGGGATTGTGGGGCTGTGTGGCCTCATATCCTC 960
D  L  F  I  P  T  G  Q  T  R  R  L  A  L  W  P  W  D  C  G  A  V  W  P  H  I  L
 T  F  S  S  P  L  D  K  L  G  V  W  R  C  G  P  G  I  V  G  L  C  G  L  I  S  S
  P  F  H  P  H  W  T  N  .  A  S  G  V  V  A  L  G  L  W  G  C  V  A  S  Y  P

CATTCTGTCTATTCTCACCCTAATCTGTCCCTGGNTACGACTCAAGCCCYGACTGACAHTGTGGTACAAGATAAGGAGGG 1040
 H  S  V  Y  S  H  P  N  L  S  L  ?  T  T  Q  A  ?  T  D  ?  V  V  Q  D  K  E  C
  I  L  S  I  L  T  L  I  C  P  W  ?  R  L  K  P  ?  L  T  ?  W  Y  K  I  R  R
P  F  C  L  F  S  P  .  S  V  P  G  Y  D  S  S  P  D  .  ?  C  G  T  R  .  G  G

AGCCCAGGTGGGTGAGATGGAAGCTGAGATGGTNCACTGTGTGCCMACCTCATTGTAATTCAACTNCCTTGACTGAAGTT 1120
 A  Q  V  G  E  M  E  A  E  M  V  H  C  V  P  T  S  L  .  F  N  ?  L  D  .  S
E  P  R  W  V  R  W  K  L  R  W  ?  T  V  C  ?  P  H  C  N  S  T  ?  L  T  E  V
 S  P  G  G  .  D  G  S  .  D  G  ?  L  C  A  ?  L  I  V  I  Q  L  P  .  L  K  L

AAAATCCAGATCCYTAGGGATGAGGGGAAGAACCTGCCAAAGACGGGTCAGGAAGGCAGTGCTAAGGGAAGGCTCCTGCA 1200
.  N  P  D  P  .  G  .  G  E  E  P  A  K  D  G  S  G  R  Q  C  .  G  K  A  P  A
 K  I  Q  I  ?  R  D  E  G  K  N  L  P  K  T  G  Q  E  G  S  A  K  G  R  L  L  Q
  K  S  R  S  L  G  M  R  G  R  T  C  Q  R  R  V  R  K  A  V  L  R  E  G  S  C

GGCCTCTGCAGTTGGACTTCATTCAGTCCCATTGCCAGAATCTCATAGCTCTTCCCYYTATCTCTCTGTCTTGAGTCTAG 1280
 G  L  C  S  W  T  S  F  S  P  I  A  R  I  S  .  L  F  P  L  S  L  C  L  E  S  S
  A  S  A  V  G  L  H  S  V  P  L  P  E  S  H  S  S  S  ?  Y  L  S  V  L  S  L
R  P  L  Q  L  D  F  I  Q  S  H  C  Q  N  L  I  A  L  P  ?  I  S  L  S  .  V  .

TTAAGAATTTGTTACCGGAGACAGAATTCTCTTTCTTAGCCTCCTGGCCAGATATTTAAAAGGAGGGGGGTGGGTTACTT 1360
 .  E  F  V  T  G  D  R  I  L  F  L  S  L  L  A  R  Y  L  K  G  G  G  W  V  T
V  K  N  L  L  P  E  T  E  F  S  F  L  A  S  W  P  D  I  .  K  E  G  G  G  L  L
 L  R  I  C  Y  R  R  Q  N  S  L  S  .  P  P  G  Q  I  F  K  R  R  G  V  G  Y  F
```

FIGURE 15a

```
TTTGGTAGGGGAAGCTTAAGTTATGGATAGCAAAGTGCTAATTGTATTCTTTTTTTCTGAAACCTCATGTAGCATTTTTC 1440
 F W . G K L K L W I A K C . L Y S F F L K P H V A F F
 F G R G S L S Y G . Q S A N C I L F F . N L M . H F S
 L V G E A . V M D S K V L I V F F F S E T S C S I F

TTCCCTTCCACCCTCCATACTTTCCCAGGCTTCATTTCATGCCCGGCGTCTCTTCGCTCACACCGCTGCAGGCTGTTTGA 1520
 F P S T L H T F P G F I S C P A S L R S H R C R L F E
 S L P P S I L S Q A S F H A R R L F A H T A A G C L
L P F H P P Y F P R L H F M P G V S S L T P L Q A V .

GGCTTCTCCCCTGGGTCTGCCTCAGCAGACTGCCTCCACACTTTCCAGTTTCTGCGTACACGTTGATATTAGAGTTTCCT 1600
 A S P L G L P Q Q T A S T L S S F C V H V D I R V S
R L L P W V C L S R L P P H F P V S A Y T L I L E F P
 G F S P G S A S A D C L R T F Q F L R T R . Y . S F L

TCCCCACTTGGCTCTTGCTCTTTCTCTGACTACCCAGGCTGATGCCATGTCTGGCCTCTTCCTGTAAATACTGTACAATG 1680
F P T W L L L F L . L P R L M P C L A S S C K Y C T M
 S P L G S C S F S D Y P G . C H V W P L P V N T V Q .
 P H L A L A L S L T T Q A D A M S G L F L . I L Y N

ATTCTATGTAAATAACTGGTCCTTGCCCACAGAGCAAGCAAGCCTTCTAGGCTAACAAATTAAAGATCAAGTTTGCTCAC 1760
 I L C K . L V L A H R A S K P S R L T N . R S S L L T
 F Y V N N W S L P T E Q A S L L G . Q I K D Q V C S
D S M . I T G P C P Q S K Q A F . A N K L K I K F A H

TGACTTTTTTATTCAATTCAAGATGGCGGGGGGTGGGGTGGGGGGGCGGATTGCCTGTTTTCACTGTGGTACCTAGGCAG 1840
 D F F I Q F K M A G G G V G G R I A C F H C G T . A
L T F L F N S R W R G V G W G G G L P V F T V V P R Q
 . L F Y S I Q D G G G W G G G A D C L F S L W Y L G R

GGCTGAAGCTCTGAGCTCCCCTGCTTTAGGCTTCTGAGTAGCCTACAGTGAGTGTTACTGTGTCCAGCTGCTCGTTGACA 1920
G L K L . A P L L . A S E . P T V S V T V S S C S L T
 G . S S E L P C F R L L S S L Q . V L L C P A A R . H
 A E A L S S P A L G F . V A Y S E C Y C V Q L L V D

TCTGGTCTCTCATGGTCTGGTCATTGTAAGCCTTAGCTCTCTGACTGTGGATGGCTTTCCTTGGCGTTAGCAGCTAACAT 2000
 S G L S W S G H C K P . L S D C G W L S L A L A A N M
 L V S H G L V I V S L S S L T V D G F P W R . Q L T
I W S L M V W S L . A L A L . L W M A F L G V S S . H

GGTTACAGGATTTCACTGAAAATTTAAATGTTGGGGGAAAGGTGCGGACACACCATAATGGTCCCAATTCAAAACAATCC 2080
 V T G F H . K F K C W G K G A D T P . W S Q F K T I
W L Q D F T E N L N V G G K V R T H H N G P N S K Q S
 G Y R I S L K I . M L G E R C G H T I M V P I Q N N P

GTGAAACAGCCTCAAGTTAGGGGTGACATGTTTTCAACCAAAGTAATTATCTTGACACCACAAAGCACACCTGTCTACAG 2160
R E T A S S . G . D V F N Q S N Y L D T T K H T C L Q
 V K Q P Q V R G E M F S T K V I I L T P Q S T P V Y R
 . N S L K L G V R C F Q P K . L S . H H K A H L S T

GCAGTGACTCCCCAAAAGCTATTAGACACACAACAAGCATGACCATAACTCAGTGGATTGGCAAGGTCACACAGTAGGAC 2240
 A V T P Q K L L D T Q Q A . P . L S Q L A R S H S R T
 Q . L P K S Y . T H N K H D H N S V D W Q G H T V G
G S D S P K A I R H T T S M T I T Q W I G K V T Q . D

TGCCCTTCACACAGTAGGTAGGAAAATGCTGCTGTCACTGCTGTCAGCTGTTATTTTGCATATCCCATGTTAAGATTAAT 2320
 A L H T V G R K M L L S L L S A V I L H I P C . D .
L P F T Q . V G K C C C H C C Q L L F C I S H V K I N
 C P S H S R . E N A A V T A V S C Y F A Y P M L R L I

AAGGCAAAAAATATTGTCTCTAAGTCCTACTTTCTGTTCCAAACTGGAGGAAATTATTGAATAAATAAACCGTGCATAAA 2400
. G K K Y C L . V L L S V P N W R K L L N K . T V H K
 K A K N I V S K S Y F L F Q T G G N Y . I N K P C I K
 R Q K I L S L S P T F C S K L E E I I E . I N R A .

AGTAGCCTCAGAAAGGGTCAAAATTTGTGTTTTCTTTGAATATTAGCTGAGGCCTCCAGGGGGCAGCACCAAGGTAGAGA 2480
 S S L R K G Q N L C F L . I L A E A S R G Q H Q G R E
 V A S E R V K I C V F F E Y . L R P P G G S T K V E
K . P Q K G S K F V F S L N I S . G L Q G A A P R . R

GCTGGACTAAGGCTGCTCTGTGTTCCTGTCCTGGGCTCCCCACAGCTCCCTTCCACCACCACTCCCATTCCATCCAACTT 2560
 L D . G C S V F L S W A P H S S L P P P L F F H P T
S W T K A A L C S C P G L P T A P F H H H S H S I Q L
 A G L R L L C V P V L G S P Q L P S T T T P I P S N F

TATTTTTAGCTGCCAGTGGGAGGGGGCAGGATAGGAGGGAAAGTAACGAAAACAGCCAAGGAGAGGGACAGAGCAACTCA 2640
L F L A A S G R G Q D R R E S N E N S Q G E G Q S N S
 Y F . L P V G G G R I G G K V T K T A K E R D R A T Q
 I F S C Q W E G A G . E G K . R K Q P R R G T E Q L

GAGCCTCTCGGACTGGACCGGACAAGCGCCCATGGAGTCTCTCTCCATCCCTCACCTGCTCCTGCCCCTGGCGTWGCTGA 2720
 E P L G L D R T S A H G V S L H P S P A P A P G V A D
 S L S D W T G Q A P M E S L S I P H L L L P L A ? L
R A S R T G P D K R P W S L S P S L T C S C P W R ? .
```

FIGURE 15b

CAGGTGAGGGAAGCAAACTTGGTTTCTGCTGGGAATGGAAGTTATGTGGATTGTTTATAATTGGGACCATTATGGCTAAA 2800
R . G K Q T W F L L G M E V M W I V Y N W D H Y G .
I C E G S K L G F C W E W K L C G L F I I G T I M A K
Q V R E A N L V S A G N G S Y V D C L . L G P L W L K

ATCTYGCGGGGCGCTCAGGTCGGAGGTTAATACCGATGCTATATTTCCTGTGTGCACTCATGTTCTTAGACACCCAAATGG 2880
N L A G A Q V G G . Y R C Y I S C V H S C S . T P K W
I ? R A L R S E V N T D A I F P V C T H V L R H P N G
S ? G R S G R R L I P M L Y F L C A L M F L D T Q M

CAGTGGCCAAAACTTCCTCTGGCTTGTACCTCATTATCTAAACCTTTGTACCTAATTATCTAAAACCTTGGTCCTAAACT 2960
Q W P K L P L A C T S L S K P L Y L I I . N L G P K L
S G Q N F L W L V P H Y L N L C T . L S K T L V L N
A V A K T S S G L Y L I I . T F V P N Y L K P W S . T

CCACAGACATGAGGGCACAGAAAAGAGACGTGTCTCTCATCTTCCATTCGGTTACACTGATTCCTACCTTCCCTGCTTCT 3040
H R H E G T E K R R V S H L P F G Y T D S Y L P C F
S T D M R A Q K R D V S L I F H S V T L I P T F P A S
P Q T . G H R K E T C L S S S I R L H . F L P S L L L

CCCTGCCATTGGTGCTCCTTGGTGCCTGAGGCATAATTGCCTTACTATGTGGTCAGAACTCTGGGTTCGCCTAACGACCG 3120
S L P L V L L G A . G I I A L L C G Q N S G F A . R P
P C H W C S L V P E A . L P Y Y V V R T L G S P N D R
P A I G A P W C L R H N C L T M W S E L W V R L T T

AGCTACAGTTTCTGGTCTCATAGCCCTGCCAATTTCCTGGATTAAAAAAAAAAGGCTCACATATAAAATACCTTTTCTGA 3200
S Y S F W S H S P A N F L D . K K K A H I . N T F S E
A T V S G L I A L P I S W I K K K R L T Y K I P F L
S L Q F L V S . P C Q F P G L K K K G S H I K Y L F .

AAATGAGCACAGTGTGAGTTGAAGTTAGATTTTGGGGATGGAGGGTTGCTTGGATGCAAAGAGCAAGACAGTAGAGAAG 3280
N E H S V S . S . I L G D G G L L G C K E Q D S R E
K M S T V . V E V R F W G H E G C L D A K S K T V E K
K . A Q C E L K L D F G G W R V A W M Q R A R Q . R R

AGAATCATGGGAGGGATAAGAGGCTGGAATTTTTCCCTGCTAGTGCCCTATAATCTTTCTTTCCTAAAATAACAGCTCTG 3360
E N H G R D K R L E F F P A S A L . S L F P K I T A L
R I M G G I R G W N F S L L V P Y N L C F L K . Q L .
E S W E G . E A G I F P C . C P I I F V S . N N S S

ATTTTATGGGAATTGGGGTCAGGAGAAAGGAATCAGTAGGCACAGATGGGACCCCAAGCGTGGACTAAAGTTTGAGGAAA 3440
I L W E L G S G E R N Q . A Q M G P Q A W T K V . G N
F Y G N W G Q E K G I S R H R W D P K R G L K F E E
D F M G I G V R R K E S V G T D G T P S V D . S L R K

CTATGGGAGTAGGCAAGGGGTGTTTGTAAGGTGGATGAGATGAGGAGATTGTGGTGGGGGGAGTCTTCGGGGTGATAGG 3520
Y G S R Q G V F V R W M R . G D C G G G E S W G . .
T M G V G K G C L . G G . D E E I V V G G S L G G D R
L W E . A R G V C K V D E M R R L W W G G V L G V I G

ACCCTTAACAGGGATAGATGGCAAACTGTGTGTGGGCAGGCCGGTGGTTCCACCCACTTAATTAGCGTTGAGGTTGGCAG 3600
D P . Q G . M A N C V W A G R W F H P L N . R . G W Q
T L N R D R W Q T V C G Q A G G S T H L I S V E V G R
P L T G I D G K L C V G R P V V P P T . L A L R L A

GGCTGGAAGGAGCCAGCACTCTCAACCTTGGAGAAAGTGCAAGTGTGACAAGAAGAAACAGAAAGAGGAGACACCCGGGC 3680
G W K E P A L S T L E K V Q V . Q E E T E R G D T R A
A G R S Q H S Q P W R K C K C D K K K Q K E E T P G
G L E G A S T L N L G E S A S V T R R N R K R R H P G

AGGGAGCTCCTTGCCATCGTTTCTTCCCATGGCCCTGGCTTTGGGAAGAATTAGGAAAGGGTGGTGACTCTGCATCCTCA 3760
G S S L P S F L P M A L A L G R I R K G W . L C I L
Q G A P C H R F F P W P W L W E E L G K G G D S A S S
R E L L A I V S S H G P G F G K N . E R V V T L H P Q

GAAAAGCCCTCTCTCCCTCTTTGGACTCTCGAGGCTTAGAGAGGAGAATGTGTAGGAGGAATGATGTGGAAAGAGTAACT 3840
R K A L S P S L D S R G L E R R M C R R N D V E R V T
E K P S L P L W T L E A . R G E C V G G M M W K E . L
K S P L S L F G L S R L R E E N V . E E . C G K S N

TGACCTATCCAGATGTGTCTGTGAATGAGATTTCAGGAATGAGAATGGAAATACAGCTGTGCTTCAGCATGGCCGAGGGC 3920
. P I Q M C L . M R F Q E . E W K Y S C A S A W P R A
D L S R C V C E . D F R N E N G N T A V L Q H G R G
L T Y P D V S V N E I S G M R M E I Q L C F S M A E G

CTTAGGATCCCTCACCCCCACCCCACAGGAAGAGAATCATCCAATCATCCCACCTGGGGTTCTGAGGACATGACATTGAC 4000
L G S L T P T P Q E E N H P I I P P G V L R T . H .
P . D P S P P P H R K R I I Q S S H L G F . G H D I D
L R I P H P H P T G R E S S N H P T W G S E D M T L T

ACAGAGCAGGAGAGCTGAGATAGAAACACTCCCTCCTGTCTTGTCTCCCACTAAGCCTCACCAGTCCTTCATTAACTGAT 4080
H R A G E L R . K H S L L S C L P L S L T S P S L T D
T E Q E S . D R N T P S C L V S H . A S P V L H . L I
Q S R R A E I E T L P P V L S P T K P H Q S F I N .

FIGURE 15c

TGGTGGATGCTAATTATGATCCTCACCCCTCAGGTCTCTGCTCCCCCTTTAATCTGGATGAACACCACCCACGACTCTTC 4160
W W M L I M I L T P Q V S A P P L I W M N T T H D S S
G G C . L . S S P L R S L L P L . S G . T P P T T L
L V D A N Y D P H P S G L C S P P N L D E H H P R L F

ACAGGGCCACCAGAGGCCGAATTTGGATACAGTGTCTTACAGCATGTTGGGGGTGGACAGCGATGGTGAGAGGGAAAACA 4240
Q G H Q R P N L D T V S Y S M L G V D S D G E R E N
H R A T R G R I W I Q C L T A C W G W T A M V R G K T
T G P P E A E F G Y S V L Q H V G G G Q R W . E G K Q

GAGGACCGTGGGATCGGGACTATGCACTCACTGATAAAGGGGAGGACCGGTCCAAGCTGGCCTTTGAAAGTGCCTGGGGC 4320
R G P W D R D Y A L T D K G E D R S K L A F E S A W G
E D R G I G T M H S L I K G R T G P S W P L K V P G A
R T V G S G L C T H . . R G G P V Q A G L . K C L G

TCCATGACGTCTCATGCACTCTCCCTCTCACTATACTAAGGACCATGCTCACCGGATCTTTATATCCATATTCTCCTTCC 4400
S M T S H A L S L S L Y . G P C S P D L Y I H I L L P
P . R L M H S P S H Y T K D H A H R I F I S I F S F
L H D V S C T L P L T I L R T M L T G S L Y P Y S P S

AGGATGCTGGTGGGTGCCCCCTGGGATGGGCCATCAGGTGACCGGAGAGGGGATGTTTATCGTTGCTCTATAGGGGGATT 4480
G C W W V P P G M G H Q V T G E G M F I V A L . G D
Q D A G G C P L G W A I R . P E R G C L S L L Y R G I
R M L V G A P W D G P S G D R R G D V Y R C S I G G F

CCACAGTGCTCCATGTACCAAAGGCCACCTGGGTAAGAAGAAGCCTGACCTTTCCCCTGCTAATTCCTGATGTTGACATC 4560
S T V L H V P K A T W V R R S L T F P L L I P D V D I
P Q C S M Y Q R P P G . E E A . P F P C . F L M L T S
H S A P C T K G H L G K K K P D L S P A N S . C . H

TAGTAACTCTGACCCCTTGGACCTTGTCTTCAATGACCCTGAACTAAAGAAGCCGAACTATGACCCCATGACTTCATTCT 4640
. . L . P L G P C L Q . P . T K E A E L . P H D F I L
S N S D P L D L V F N D P E L K K P N Y D P M T S F
L V T L T P W T L S S M T L N . R S R T M T P . L H S

CTTCTACCCTTCCTCCAACCAGGTGACTATCAACTTGGAAATTCCTCTCAGCCTGCTGTGAATATGCACCTAGGGATGTC 4720
F Y P S S N Q V T I N L E I P L S L L . I C T . G C
S S T L P P T R . L S T W K F L S A C C E Y A P R D V
L L P F L Q P G D Y Q L G N S S Q P A V N M H L G M S

TCTACTAGAGACAGATGCTGATGGGGATTCATGGTGAGCTGAAAGAAGGGCCTCAGAAGGTTCACAGCAGGGAAGAGAG 4800
L Y . R Q M L M G D S W . A E R R A S E G S Q Q G R E
S T R D R C . W G I H G E L K E G P Q K V H S R E E S
L L E T D A D G G F M V S . K K G L R R F T A G K R

CATTATGGTATCTGGGCAGTGGTGGCTTGGGCCTTTCATCCCAGTGTTCTGGAGGCAGAGTCAGGCCTGATCTACAGAGT 4880
H Y G I W A V V A W A F H P S V L E A E S G L I Y R V
I M V S G Q W W L G P F I P V F W R Q S Q A . S T E
A L W Y L G S G G L G L S S Q C S G G R V R P D L Q S

GAGCTCCAGGACAGCCAAGGCTATGCAGAGAAACCCTGTTTTGAAAAACCCAAAACCAAAACTAACCAAACAACAACAAC 4960
S S R T A K A M Q R N P V L K N P K P K L T K Q Q Q
. A P G Q P R L C R E T L F . K T Q N Q N . P N N N N
E L Q D S Q G Y A E K P C F E K P K T K T N Q T T T T

AGAAAAAGCACCGTGGTAAGGGAAATTAGTCTGTATAGAAGAGACAAGGAATTCAAAACCCTAGAGAGCAAGGCAGGGTT 5040
Q K K H R G K G N . S V . K R Q G I Q N P R E Q G R V
R K S T V V R E I S L Y R R D K E F K T L E S K A G F
E K A P W . G K L V C I E E T R N S K F . R A R Q G

CCCCATGGAGTGGTCTCCATCTCTCTTTTAACTAGGTGTGTGTTCCGAGAGGCCCTCTCAAGCCTGGGGATAACTATTTC 5120
P H G V V S I S L L T R C V F R E A L S S L G I T I S
P M E W S P S L F . L G V C S E R P S Q A W G . L F
S P W S G L H L S F N . V C V P R G P L K P G D N Y F

TCCTATCCACCCAGGCCTGTGCCCCTCTTTGGTCTCGTGCCTGCGGCAGCTCTGTCTTCAGTTCTGGAATATGTGCCCGT 5200
P I H P G L C P S L V S C L R Q L C L Q F W N M C P
L L S T Q A C A P L W S R A C G S S V F S S G I C A R
S Y P P R P V P L F G L V P A A A L S S V L E Y V P V

GTGGATGCTTCATTCCGGCCCCAGGGAAGCCTGGCACCCACCGCCCAACGTGAGCCAGTGGAAGGGCCCTGGAAGCTCAG 5280
C G C F I P A P G K P G T H R P T . A S G R A L E A Q
V D A S F R P Q G S L A P T A Q R E P V E G P W K L S
W M L H S G P R E A W H P P P N V S Q W K G P G S S

TTCCCAGATAGGGATGCTGGGTGGGAAAAACTAGGACAAAGACTTGGTGGAGGGTCTGCATGGCTATCCTCATCATTCCC 5360
F P D R D A G W E K L G Q R L G G G S A W L S S S F P
S Q I G M L G G K N . D K D L V E G L H G Y P H H S
V P R . G C W V G K T R T K T W W R V C M A I L I I P

AAGTGTGCTTGCAGAAGAGGCTCCTGTTTGCTAACTGATTAGAATTCAGACTCCTTAGGAGAGCCTCAAGACACCAGGAT 5440
S V L A E E A P V C . L I R I Q T P . E S L K T P G
Q V C L Q K R L L F A N . L E F R L L R R A S R H Q D
K C A C R R G S C L L T D . N S D S L G E P Q D T R I

FIGURE 15d

CTGGTTTTACCAACTTAAAAACAAAACAAAACAGCATATCCTGTGCACAGCCTATCCCTCATCCATCACGTGTCCTCCAT 5520

S G F T N L K T K Q N S I S C A Q P I P H P S R V L H
L V L P T . K Q N K T A Y P V H S L S L I E H V S S I
W F Y Q L K N K T K Q H I L C T A Y P S S I T C P P

ATCTTATTTTTGTGGGTCTTATAGATGCCAAGTCAGCACTCAGTTATTGGGTTCTCCCCTCATGCCTTTCATATACTTTC 5600

I L F L W V L . M P S Q H S V I G F S P H A F H I L S
S Y F C G S Y R C Q V S T Q L L G S P L M P F I Y F
Y L I F V G L I D A K S A L S Y W V L P S C L S Y T F

TTATCTACTGCCTTTTGGGAGATAGTCTTATGTAGCCCAGGCTGTCCTTGATCTTGGAATTTGCTTGCCTCAGCTTCTCA 5680

Y L L P F G R . S Y V A Q A V L D L G I C L P Q L L
L I Y C L L G D S L M . P R L S L I L E F A C L S F S
L S T A F W E I V L C S P G C P . S W N L L A S A S Q

GTCTCAAGTACTGGGATAATAGGCATGCATTGTCTGCCTGGCCTTTGCTGAACATGCCCTCTGTGGCCATTGGTAGGGCA 5760

S L K Y W D N R H A L S A W P L L N M P S V A I G R A
V S S T G I I G M H C L P G L C . T C P L W P L V G H
S Q V L G . . A C I V C L A F A E H A L C G H W . G

TGAGTCAAATACTGCCCTCCCCCACAACACACACACAAACGAAAGTGAGGCTCTCTAAGTGTTCCATAGCACAGGGTAGT 5840

. V K Y C P P P Q H T H K R K . G S L S V P . H R V V
E S N T A L P H N T H T N E S E A L . V F H S T G .
M S Q I L P S P T T H T Q T K V R L S K C S I A Q G S

GGTAGGCCTCTCGCTAGTGCATATTTCATTCTTTTACTCTGCCCATCTCTTCTTTCTTTGATTTCCACACTGGGGACCTG 5920

V G L S L V H I S F F Y S A H L F F L . F P H W G P
W . A S R . C I F H S F T L P I S S F F D F H T G D L
G R P L A S A Y F I L L L C P S L L S L I S T L G T W

GCATAGTACTTTCCTGGTAATTAAGAGAGAATTCCCTTTTAAGTGCCTGCATTGCAGCGTCCTCCTGGGACATTCTCCCT 6000

G I V L S W . L R E N S L L S A C I A A S S W D I L P
A . Y F P G N . E R I P F . V P A L Q R P P G T F S L
H S T F L V I K R E F P F K C L H C S V L L G H S P

TGCTGACTACACCCCACATCCTTCCATGTTTTTTGTTTCCCATCACTATGCCCCCCTTCTAGGCTGTCCCACATACATGG 6080

C . L H P T S F H V F C F P S L C P P S R L S H I H G
A D Y T P H P S M F F V S H H Y A P L L G C P T Y M
L L T T P H I L P C F L F P I T M P P F . A V P H T W

ATGTCGTCATTGTTTTGGATGGCTCCAACAGTATCTATCCCTGGTCAGAAGTTCAGACTTTCCTTCGGAGGCTGGTAGGA 6160

C R H C F G W L Q Q Y L S L V R S S D F P S E A G R
D V V I V L D G S N S I Y P W S E V Q T F L R R L V G
M S S L F W M A P T V S I P G Q K F R L S F G G W . E

AGACTGTTCATCGATCCGGAGCAGATACAGGTAAGAGAAAGATATGTGGATAGGATTGGAGGGAAAGAAGTAAACACTCC 6240

K T V H R S G A D T G K R K I C G . D W R E R S K H S
R L F I D P E Q I Q V R E R Y V D R I G G K E V N T P
D C S S I R S R Y R . E K D M W I G L E G K K . T L

TGGACCCTTGGATGTAAGCAGCCATGTCCAGCCTCTTGATGACACCCTGGGACATTGTCTTCTACAGAACTCATGCTCAA 6320

W T L G C K Q P C P A S . . H P G T L S S T E L M L K
G P L D V S S H V Q P L D D T L G H C L L Q N S C S
L D P W M . A A H S S L L M T P W D I V F Y R T H A Q

GAACTGTGCAATTAACTTACCAAAAAGTCACAAAAATTTCATAATGTTTGAAGTAAGTTTATGATTGTGTGGGGGGCCAC 6400

N C A I N L P K S H K N F I M F E V S L . L C G G P
R T V Q L T Y Q K V T K I S . C L K . V Y D C V G G H
E L C N . L T K K S Q K F H N V . S K F M I V W G A T

ACTCAGAGCTTCCCTTTGCTGCTTGTAGTTGCTTGGGCAATGCATGCCATGAGCTGCAAGTTAGACACACCTGTTCACTT 6480

H S E L P F A A C S C L G N A C H E L Q V R H T C S L
T Q S F P L L L V V A W A M H A M S C K L D T P V H F
L R A S L C C L . L L G Q C M P . A A S . T H L F T

CCCCTTCATCGTGCTGCAGGTTGGACACACCTGTTAGGGGTTCACTTCCCCTTCATCCTTTGTGCTCCATCTTCTCTACG 6560

P L H R A A G W T H L L G V H F P F I L C A P S S L R
P F I V L Q V G H T C . G F T S P S S F V L H L L Y
S P S S C C R L D T P V R G S L P L M P L C S I F S T

CTCTTCATACATCCCATGTGGGCACATGGTCTATTGTTCTCAGGTAGGACTGGTACAGTACGGGGAGAACCCTGTGCATG 6640

S S Y I P C G H M V Y C S Q V G L V Q Y G E N P V H
A L H T S H V G T W S I V L R . D W Y S T G R T L C M
L F I H P M W A H G L L F S G R T G T V R G E P C A .

AGTGGTCCCTGGGAGACTTCCGAACAAAGGAAGAAGTTGTGAGAGCAGCAAGGAACCTAAGTCGGAGGGAAGGGCGAGAA 6720

E W S L G D F R T K E E V V R A A R N L S R R E G R E
S G P W E T S E Q R K K L . E Q Q G T . V G G K G E K
V V P G R L P N K G R S C E S S K E P K S E G R A R

ACGAGAACCGCCCAAGCGATCATGGTGGCATGGTGAGACATTGTAAAGGGGTCGTGTGAGGGAGGAGGAAGGATCAGCAG 6800

T R T A Q A I M V A W . D I V K G S C E G G G R I S R
R E P P K R S W W H G E T L . R G R V R E E E G S A
N E N R P S D H G G M V R H C K G V V . G R R K D Q Q

FIGURE 15e

GGAGAGGGAGAGGGTCTGGAGTGTAGTGTATACATCACAAGATGCTCTGGGCGCTTATCTTTATCTGCATGCCAGAAGTT 6880

E R E R V W S V V Y T S Q D A L G A Y L Y L H A R S
G R G R G S G V . C I H H K M L W A L I F I C M P E V
G E G E G L E C S V Y I T R C S G R L S L S A C Q K F

CGTGGAGGAAGGCTAGGTTGCTGTCACCATACTCTCTCTTACTGTATTTGCATTTTATGGTGTCTGTGGGTGTATCTCTC 6960

S W R K A R L L S P Y S L L L Y L H F M V S V G V S L
R G G R L G C C H H T L S Y C I C I L W C L W V Y L S
V E E G . V A V T I L S L T V F A F Y G V C G C I S

CTTGTCTGTTCTGTTTCTGCACACAGAACTCCATCTTTCCTCTTCTACTCCTGCGTCAATTCTGATACCTAGCTTCTCAA 7040

L V C S V S A H R T P S F L F Y S C V N S D T . L L N
L S V L F L H T E L H L S S S T P A S I L I P S F S
P C L F C F C T Q N S I F P L L L L R Q F . Y L A S Q

CCACTCACGCCCTAGTATTCTTTTCAAACATGACTCTAAACCTCTGGGGAGGCTACATGACCTGACTGTCTTTATTCTCC 7120

H S R P S I L F K H D S K P L G R L H D L T V F I L
T T H A L V F F S N M T L N L W G G Y M T . L S L F S
P L T P . Y S F Q T . L . T S G E A T . P D C L Y S P

AGTTCCTTGATCTTGTCAACCCAAGTGTTTGCTGAATGAATCTATAAATAAATAATGCTTGTACATATTTACACTGATGA 7200

Q F L D L V N P S V C . M N L . I N N A C T Y L H . .
S S L I L S T Q V F A E . I Y K . I M L V H I Y T D D
V P . S C Q P K C L L N E S I N K . C L Y I F T L M

CAGATTATTTTATATGTTCCGTGCCATCTAAACAGTCAAGTTGTGACTCTGTGCCAGTTTGCATGCTAGATACTGTTGGG 7280

Q I I L Y V P C H L N S Q V V T L C Q F A C . I L L G
R L F Y M F R A I . T V K L . L C A S L H A R Y C W
T D Y F I C S V P S K Q S S C D S V P V C M L D T V G

GAATGGTGTAGAAGACATCTGACCTCAGTGAACTGCTGACAGTGTTAATACACTATACGGGCATGCCTGCATGCAAGCCT 7360

N G V E D I . P Q . T A D S V N T L Y G H A C M Q A
G M V . K T S D L S E L L T V L I H Y T G M P A C K P
E W C R R H L T S V N C . Q C . Y T I R A C L H A S L

GTGTGTATGTGCATGCATATGCACACACATACATATGACCATATAGCATTCTTTTATCTCTCTTCTTAGCACAGAAGGGT 7440

C V Y V H A Y A H T Y I . P Y S I L L S L F L A Q K G
V C M C M H M H T H T Y D H I A F F Y L S S . H R R V
C V C A C I C T H I H M T I . H S F I S L L S T E G

TCAGTCAGTCCCGGGGGGGACGACCAGAGGCCGCTAGGCTGCTGGTAGTTGTCACTGATGGAGAGTCCCATGATGGAGAG 7520

S V S P G G D D Q R P L G C W . L S L M E S P M M E R
Q S V P G G T T R G R . A A G S C H . W R V P . W R
E S Q S R G G R P E A A R L L V V V T D G E S H D G E

GAACTTCCAGCAGCGCTAAAGGCCTGTGAGGCTGGCAGAGTGACACGTTATGGGATTGCGGTGAGACTTGATCAAGTCCA 7600

N F Q Q R . R P V R L A E . H V M G L R . D L I K S
G T S S S A K G L . G W Q S D T L W D C G E T . S S P
E L P A A L K A C E A G R V T R Y G I A V R L D Q V Q

GTTGTTTTGTTTTGTGTTGTATCGTGTGTGTGTGTGTGTGTGTGTGTGTGTGTGTGTGTGTGTGTATGTGTGATAT 7680

S C F V L C C I V C V C V C V C V C V C V Y V . Y
V V L F C V V S C V C V C V C V C V C V C M C D M
L F C F V L Y R V C V C V C V C V C V C V C V I

GTGTGCATGCATCAGTGCACATACCATAGTGTGTATATGCGGGTCAGAGAACAACCTCAGATGTTGGTCCTCACCTTCCA 7760

V C M H Q C T Y H S V Y M R V R E Q P Q M L V L T F H
C A C I S A H T I V C I C G S E N N L R C W S S P S
C V H A S V H I P . C V Y A G Q R T T S D V G P H L P

TCTTGTTCCAAACTGGATATCTTGTTCACTTCGGCATACAATAAGCCAGATTAGCTGACCCACAAGTCTTGGGCAGGTCT 7840

L V P N W I S C S L R H T I S Q I S . P T S L G Q V
I L F Q T G Y L V H F G I Q . A R L A D P Q V L G R S
S C S K L D I L P T S A Y N K P D . L T H K S W A G L

TCTGTCTCAGCCTCCTGTCTCTTGGTTTGAGGCATTCTGGAATTTACAGATAAGCTTGATATCGAATTCCTGCAGCCCGG 7920

F C L S L L S L G L R H S G I Y R . A . Y R I P A A R
S V S A S C L L V . G I L E F T D K L D I E F L Q P G
L S Q P P V S W F E A F W N L Q I S L I S N S C S P

GGGATCCACTAGTTCTAGAGCGGCCGCCACCAAGGGAG 7958

G I H . F . S G R H Q G S
G S T S S R A A A T K G
G D P L V L E R P P P R E

FIGURE 15f

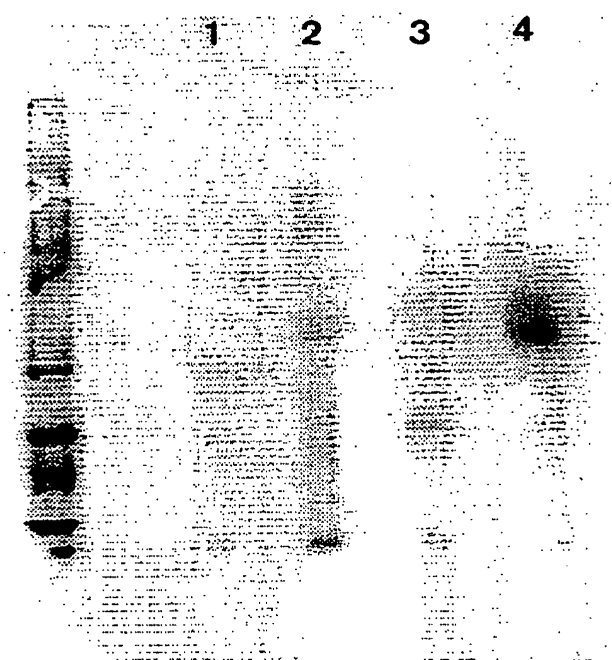


FIGURE 16

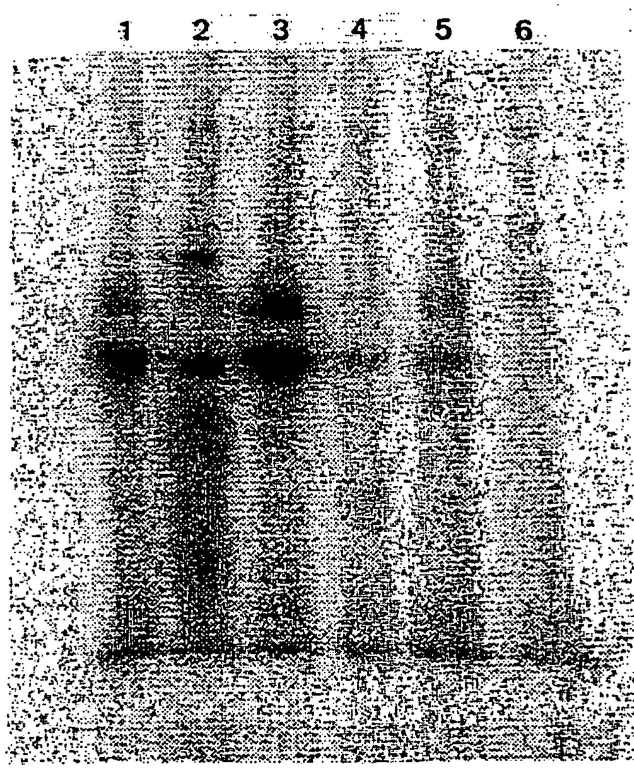


FIGURE 17

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SE 99/00544

A. CLASSIFICATION OF SUBJECT MATTER

IPC6: C07K 14/705, A61K 38/17, C07K 16/28
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC6: C07K, A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

SE,DK,FI,NO classes as above

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
P,X	J Biol Chem., Volume 273, August 1998, Lisbet Camper et al, "Isolation, Cloning, and Sequence Analysis of the Integrin Subunitalpha 10, a Betal-associated Collagen Binding Integrin Expressed on Chondrocytes", Issue32, page 20383 - page 20389	1-86
	--	
X	WO 9219647 A1 (THE SCRIPPS RESEARCH INSTITUTE), 12 November 1992 (12.11.92)	1-20,25-29, 33-44,48-53, 57-62,66-86
A		21-24,30-32, 45-47,54-56, 63-65
	--	

[X] Further documents are listed in the continuation of Box C. [X] See patent family annex.

* Special categories of cited documents:
"A" document defining the general state of the art which is not considered to be of particular relevance
"E" erlier document but published on or after the international filing date
"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
"O" document referring to an oral disclosure, use, exhibition or other means
"P" document published prior to the international filing date but later than the priority date claimed
"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"X" document of particular relevance: the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance: the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
14 July 1999	30-07-1999

Name and mailing address of the ISA/	Authorized officer
Swedish Patent Office Box 5055, S-102 42 STOCKHOLM Facsimile No. +46 8 666 02 86	Patrick Andersson/Els Telephone No. +46 8 782 25 00

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SE 99/00544

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	J Cell Biol, Volume 115, No 1, October 1991, Takada Y, Murphy et al, "Molecular cloning and expressin of the cDNA for alpha 3 subunit of human alpha 3 beta 1 (VLA-3), an integrin receptor for fibronectin, laminin, and collagen", page 257 - page 266, Medline abstract acc. no. 92011866	1-20,25-29, 33-44,48-53, 57-62,66-86
A		21-24,30-32, 45-47,54-56, 63-65
	--	
X	EMBO J, Volume 8, No 5, May 1989, Takada Y et al, "The primary structure of tha alpha 4 subunit of VLA-4: homology to other integrins and a possible cell-cell adhesion function", page 1361 - page 1368, Medline abstract Acc. no. 89356603	1-20,25-29, 33-44,48-53, 57-62,66-86
A		21-24,30-32, 45-47,54-56, 63-65
	--	
X	J.Cell Biiol., Volume 138, No 5, Sept 1997, Lisbet Camper et al, "Integrin alpha2Betal Is a Receptor for the Cartilage Matrix Protein Chondroadherin" page 1159 - page 1167	1-20,25-29, 33-44,48-53, 57-62,66-86
A		21-24,30-32, 45-47,54-56, 63-65
	--	
X	WO 9425487 A1 (CHILDREN'S MEDICAL CENTER CORPORATION), 10 November 1994 (10.11.94), page 16 - page 21	1-19,28, 33-42,48-52, 57-61,66-86
A		20-27,43-47, 53-56,62-65
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INTERNATIONAL SEARCH REPORT

International application No.
PCT/SE 99/00544

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	EP 0330506 A2 (DANA-FARBER CANCER INSTITUTE), 30 August 1989 (30.08.89) -- ---------	1-86

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SE99/00544

Box I — Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.: 28-41, 43-69, 74-79, 83-85
because they relate to subject matter not required to be searched by this Authority, namely:
These claims relate to either methods of treatment by therapy or diagnostic methods practised on the human or animal body, see PCT Rule 39.1(iv). Nevertheless, a search has been executed for these claims. The search has been based on the see next page

2. [ ] Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

Box II — Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. [ ] As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

alleged effects of the compounds.

INTERNATIONAL SEARCH REPORT

Information on patent family members

01/06/99

International application No.

PCT/SE 99/00544

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9219647 A1	12/11/92	AU 1896392 A	21/12/92
		US 5310874 A	10/05/94
		US 5589570 A	31/12/96
WO 9425487 A1	10/11/94	AU 6639394 A	21/11/94
EP 0330506 A2	30/08/89	JP 2003700 A	09/01/90
		US 5583203 A	10/12/96